Josefine Neuendorf

Urine Sediment

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Foreword

The advantages of urine diagnosis are undisputed:

- Readily obtainable test material
- Fast diagnosis
- Fast results
- High differential diagnostic reliability
- Cost-effective diagnosis

Although medical knowledge has advanced in terms of diagnostic urine sediment analysis, current knowledge is not sufficiently integrated in modern diagnostic methods. It is essential that precision be reintroduced. What is seen using microscopy can be identified in a more differentiated manner, and thus better interpreted. General statements such as "urine sediment contains epithelial cells and/or erythrocytes" are unhelpful. It is important to consider the characteristic morphological features of a urinary sediment constituent, as well as classify and name it accordingly. Only in this way is it possible to formulate a well-founded urine sediment finding suggestive of renal or post-renal disease.

The central aim of this book is to reappraise sediment diagnosis with an up-to-date naming system and an evaluation of urinary sediment constituents from a differential diagnostic perspective. Furthermore, detailed instructions on the precise method for processing urine into urine sediment are provided.

The knowledge imparted here on microscopy techniques, as well as on servicing and maintaining the microscope, is crucial for ease of work and facilitates the morphological determination of cell components. All aspects of correct urine processing discussed here guarantee reproducible results.

Autodidactic acquisition of cell morphology is highly challenging and time-consuming, since microscopy specimens are fresh native specimens that cannot be fixed and, as such, cannot be archived.

Therefore, precise photographs of the constituents of urine sediment are required in order to be able to more readily compare and classify what is seen microscopically. For this reason, the book contains a multitude of digital photographs from both bright-field microscopy and phase-contrast microscopy. Furthermore, numerous examples provide the reader with the opportunity to practice evaluating and diagnosing microscopic images.

In the context of my lecture activities, I encounter a considerable need for information on urine diagnostics among nephrologists, urologists, gynecologists, medical students, medical technical laboratory assistants, and medical specialists. The extremely positive feedback from seminar participants prompted me to compile the main contents of my lectures in book form.

Heidelberg, Germany — Josefine Neuendorf
2019

Preface

The inspection of urine has been of particular relevance in the identification of diseases for almost 2000 years. The urine glass, known as the matula, was even the status symbol for physicians in the Middle Ages. As a result of the automation of laboratory diagnostics and the introduction of urine test strips, a considerable portion of urine diagnostics can be standardized now. However, when it comes to the analysis of urine sediment, an experienced diagnostician is by far superior to automated urine sediment analysis.

In her book, Josefine Neuendorf presents all the information needed for urinary sediment analysis, from obtaining urine, pre-analytics, and analysis to urine sediment diagnosis in a clear and concise manner. Each of these analytical steps on the way to reliable diagnosis based on urine sediment are made clear and convincing thanks to the easy-to-understand presentation of the basic principles on the one hand, and the detailed explanations on differentiating morphological details on the other. The 3rd edition of the book has gained considerably from the clear depiction of urinary sediment constituents using high-resolution images of urine sediment.

Well performed urinary sediment analysis can make a valuable contribution to the differential diagnoses of acute kidney diseases, especially when the arguments for the various differential diagnoses need to be weighed up in a timely manner. Properly obtained and correctly interpreted findings of urinary sediment analysis can support the indication for kidney biopsy or the decision to initiate treatment immediately.

Questions on urine sediment are also part of exams. Thus, according to the catalog of learning targets issued by the German Institute for Medical and Pharmaceutical Examination Questions (IMPP), students should be familiar with performing urinary sediment analysis. In particular, the differentiation of glomerular from non-glomerular erythrocyturia is an important examination topic, which is tested by asking students to identify acanthocytes on images of urine sediment. Moreover, the German Specialty Training Regulations for Nephrology call for experience and skills in the performance of—and reporting of findings from — microscopic analysis of urine sediment, including phase contrast microscopy. This book provides a clear explanation of all these aspects.

Therefore, the book “Urine Sediment” represents not only a valuable aid in daily practice, but also during studies and continuing education.

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Head of the Department of Nephrology University Hospital Essen
Germany

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Abbreviations

aHPF	All (analyzed) high power fields
Acantho	Acanthocyte/s
Amph	Amorphous phosphates
Bact	Bacteria
Biconcave Ec	Biconcave-shaped erythrocyte/s
Bright field	Bright-field microscopy
Ca-Oxa	Calcium oxalates
Ca-oxalates	Calcium oxalates
cm	Centimeter
Disc-shaped Ec	Disc-shaped erythrocytes
dUc	Deep urothelial cell/s
DysEc	Dysmorphic erythrocyte/s
dysmorphic Ec	Dysmorphic erythrocyte/s
Ec	Erythrocyte/s
Ec ghosts	Erythrocyte ghosts
EcCa	Erythrocyte Cast/s
EpiCa	Epithelial cast/s
EumEc	Eumorphic erythrocyte/s
Eumorphic Ec	Eumorphic erythrocyte/s
FC	Fatty cast/s
GranCa	Granular cast/s
HPF	High power field
HyalCa	Hyaline cast/s
Lc	Leukocyte/s
LcCa	Leukocyte cast/s
mm	Millimeter
NR	Normal range
OFB	Oval fat bodies
OFB casts	Oval fat body casts
PhaCo	Phase-contrast microscopy
SqEc	Squamous epithelial cell/s
Thornapple Ec	Thorn apple-shaped erythrocyte/s
TransEc	Transitional Epithelial Cell/s
UTI	Urinary tract infection
YC	Yeast cell/s

About the Author

Josefine Neuendorf MTLA, lecturer in medical laboratory diagnostics, gives lectures and practical seminars for physicians, MTLAs, and medical staff and teaches at the Heidelberg Academy for Health Professions at the Heidelberg University Hospital, Germany.

Part I

The Microscope

1

1.1 Structure of the Microscope

Figures 1.1 and 1.2 show the magnification calculation and the structure of a microscope.

1.2 Cleaning and Maintaining the Microscope

- Vibrations should be avoided when the light source is on, since the lamp responds highly sensitively to this.
- Protect the microscope from exposure to dust, e.g., by using a dust cover/plastic cover, and by closing openings that dust could get into (eyepieces should always be pushed in). All positions on the objective turret (revolving nosepiece) must be fitted either with an objective or with a dust protection plastic plug (Fig. 1.4).

Remove dust from objectives and eyepieces by blowing or dabbing with a fine microfiber cloth and then cleaning with a high-quality paper tissue (swabs, linen cloths, cotton buds, and eyeglass cleaning cloths should not be used) soaked in rinsing solution or glass cleaner solution. Dry paper tissues should never be used.

- A good cleaning solution for all optical glass surfaces and the tripod consists of a mixture of 1 l glass cleaner and 20–30 ml odorless methylated spirits.
- A particular technique is recommended for cleaning glass lenses (Fig. 1.3):

  Only ever wipe the glass lens in a circular motion, otherwise dirt on the glass lens collects at the edge of the lens. Avoid using cotton buds, since these cause renewed smearing. Only ever clean external glass surfaces. The cleaning of internal glass surfaces can only be performed by a professional specialist.
- In order to clean the outer objective lens more effectively and thoroughly, the objective can also be unscrewed from the microscope from time to time.
- Never oil the precision guides, drive mechanisms, screws, or moving parts.
- If an objective is missing from the nosepiece, the holder must be closed with a plastic dust cap (Fig. 1.4).

1.3 Servicing the Microscope

- Depending on its use, the microscope needs to be serviced by a specialist at regular intervals. This includes checking the microscope for functionality, cleanliness, resin, etc.
- It is a legal requirement in Germany for the power cable and all electrical parts be checked by a specialist once a year. Following inspection, the electrician affixes a sticker to the microscope.

1.4 Light Bulb Replacement

- When replacing the microscope lamp, it is essential that the halogen lamp is not touched with the naked hand under any circumstances.
- A lint-free linen cloth must be used.

Magnification is calculated as follows:

10x Eyepiece · 40x objective = 400x magnification

8x Eyepiece · 40x objective = 320x magnification

Fig. 1.1 Calculating magnification

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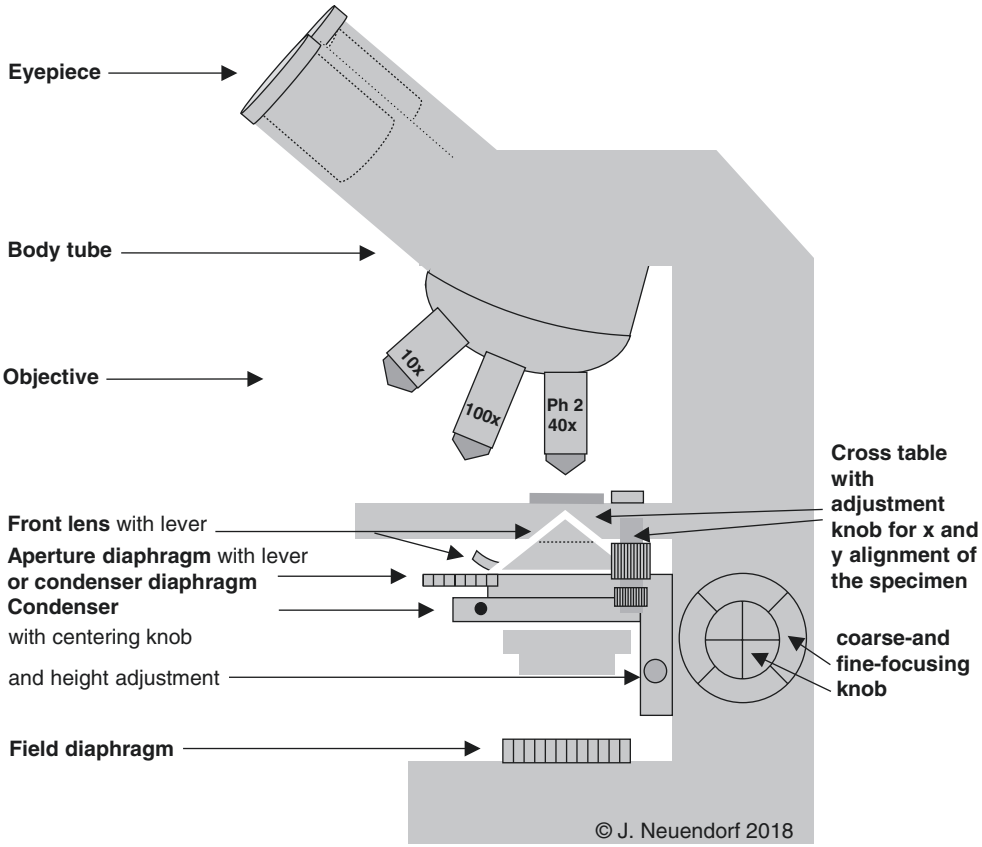


Fig. 1.2 Structure of the microscope

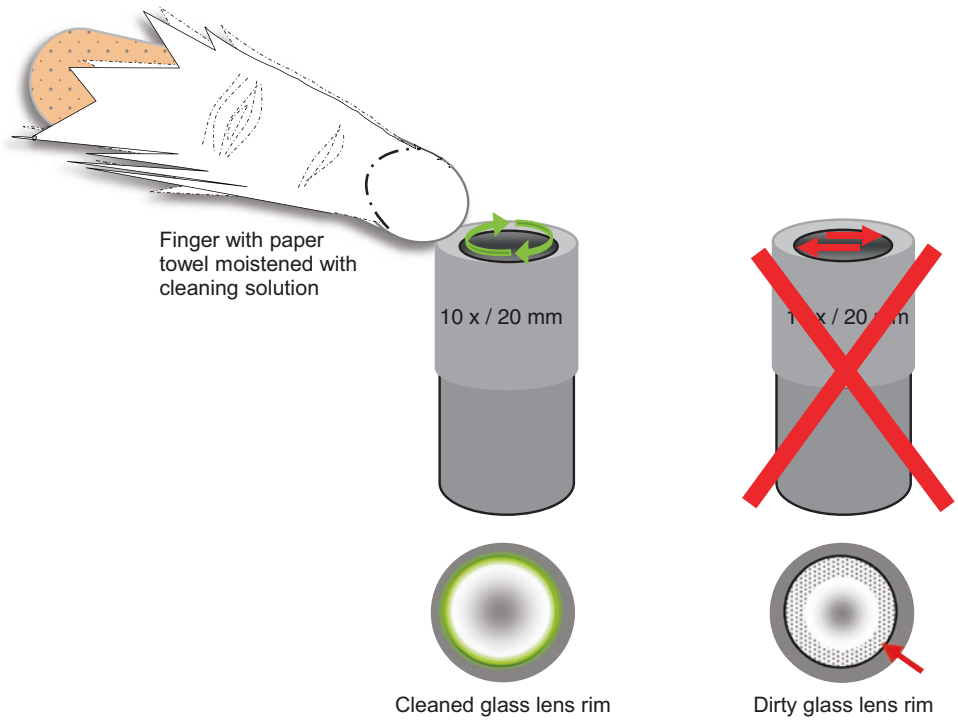


Fig. 1.3 Cleaning glass lenses

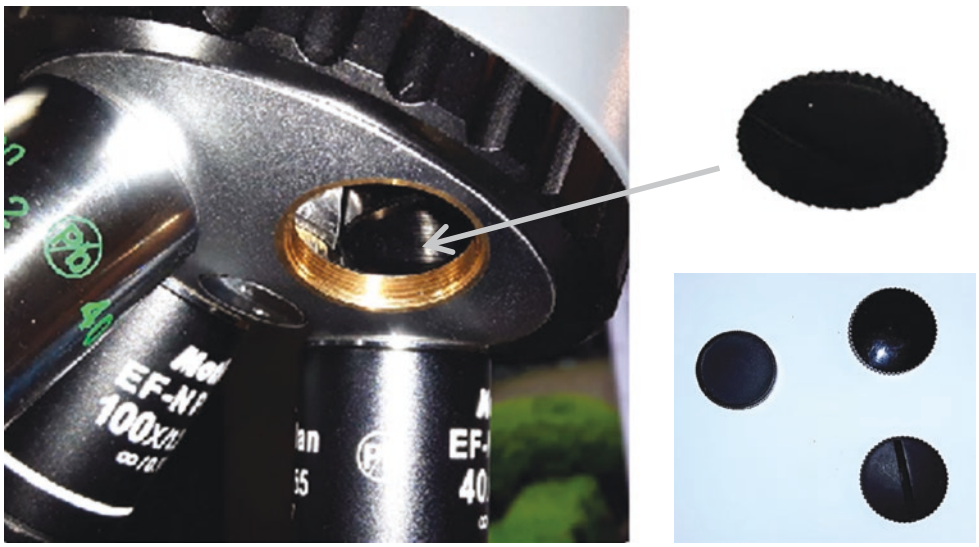


Fig. 1.4 Nosepiece without microscope plastic dust cap, various plastic caps

Setting-Up Köhler Illumination

2

2.1 Setting-Up Köhler Illumination or Aligning the Microscope

Professor August Köhler (1866–1948) worked for Carl Zeiss in Jena, Germany and, in 1893, published a set of rules for the correct illumination of microscopic specimens.

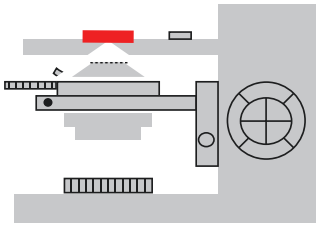
The aim here is to achieve uniform illumination of the microscopic image while at the same time increasing resolution capacity by using a condenser. Unwanted reflections and contrast-reducing overexposure are largely eliminated (Zeiss 1997).

Köhler illumination has been described for bright-field and phase-contrast microscopy (Figs. 2.1 and 2.2). In order to be able to better set the microscopic plane of the slide, one uses a stained specimen (Köhler specimen) to set-up Köhler illumination. This can be a stained blood smear from the hematology department or simply a slide marked with a felt-tip pen.

> Important: The Köhler specimen should be of the same thickness as the specimens to be subsequently examined microscopically. Attention should be paid to this if one is working with KOVA® slides/counting chambers.

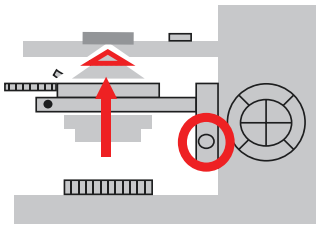
J. Neuendorf, *Urine Sediment*, https://doi.org/10.1007/978-3-030-15911-5_2

1



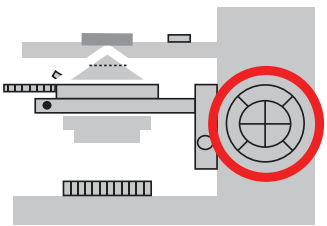
Place the slide or Köhler specimen on the stage.

2



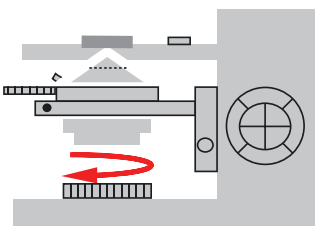
Turn the front lens (if there is one) into the beam.
Turn the condenser upwards.

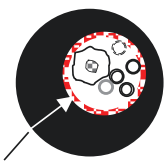
3



Using the 10x objective, bring the microscopy plane of the slide into sharp focus.

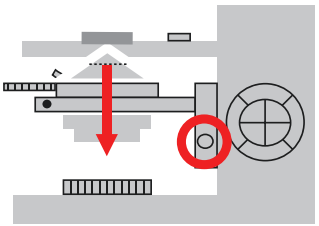
4





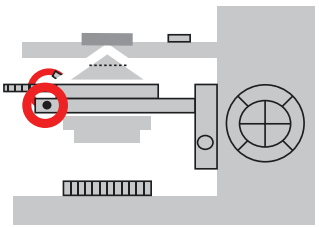
Close the field diaphragm completely while viewing. A bright circle (or hexagon) with an unsharp border will become visible against a dark background.

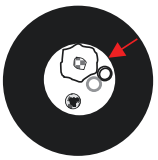
5



Lower the condenser slightly while viewing until the edge of the bright circle comes into sharp focus.

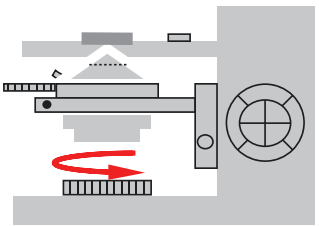
6





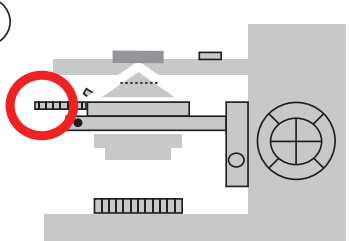
While still viewing, turn the bright circle to the center using the two condenser-centering knobs (left and right).

7



Again while viewing, open the field diaphragm only so wide that the entire field of view is illuminated; readjust slightly if necessary as described in 6.

8



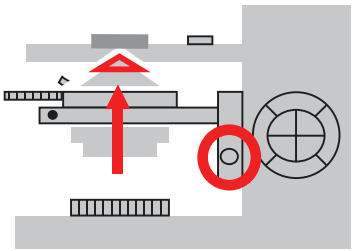
To adjust image contrast, close the aperture diaphragm lever approximately two thirds.

- Set-up Köhler illumination also using a 40x objective!

Fig. 2.1 Setting up Köhler illumination or aligning the microscope

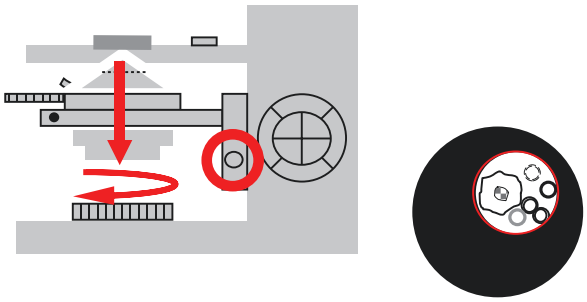
2.2 Quick Guide to Setting-Up Köhler Illumination

1. Place the Köhler specimen on the stage.
   Place the condenser on the highest setting.
   Push the condenser front lens (if there is one) away from the stage.



2. Using the 10x objective, bring the specimen into sharp focus using the coarse- and fine-focusing knob.

3. Close the field diaphragm in the foot of the microscope and slowly lower the condenser until the field diaphragm image comes into focus (hexagon or circle).



4. Turn both condenser-centering knobs until the image of the field diaphragm appears at the center of the field of view.
   The condenser is now centered.

5. While viewing, open the field diaphragm only wide enough for the entire field of view to be illuminated.
   Adjust image contrast using the aperture diaphragm (condenser diaphragm).

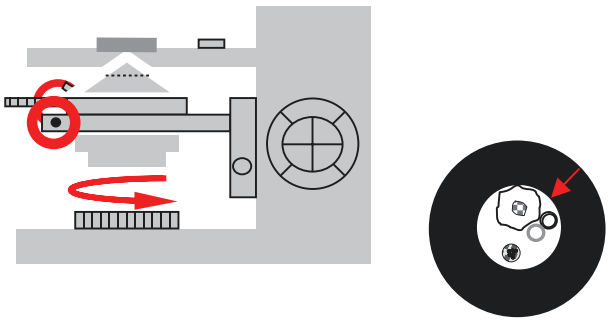


Fig. 2.2 Quick guide to Köhler illumination

Reference

1. Zeiss C (1997) Mikroskopieren von Anfang an! Was heißt Köhlern? http://www.meditec.zeiss.de/C1256B5E-00496AB1/Contents-Frame/2BEEE02E723F-BA45C1257346003FB23C?opendocument. Accessed on: 23rd January 2015.

Phase-Contrast Microscopy

3

3.1 Application

Phase-contrast microscopy is used to better examine unstained specimens, native specimens, or low-contrast elements of urine. Originally transparent structures are made visible by virtue of the fact that they are surrounded by a dark and a light border.

3.2 What Is Required for Changeover?

By swapping a special **phase-ring objective** and inserting a phase annulus in the **condenser**, a normal bright-field microscope can be rapidly converted into a phase-contrast microscope. It is important here that the phase annulus in the condenser and the phase ring in the objective are centered relative to each other. This is done using an **auxiliary microscope** or viewfinder **diopter**.

What is practical is that both microscopy techniques (bright-field and phase-contrast microscopy) can be used effortlessly in parallel.

3.3 The Light Pathway of Phase-Contrast Microscopy

See Fig. 3.1

3.4 Phase-Contrast Microscopy Equipment

This includes a PhaCo objective and various PhaCo condensers:

3.4.1 PhaCo Objective

See Fig. 3.2

3.4.2 PhaCo Condensers

See Figs. 3.3 and 3.4

J. Neuendorf, *Urine Sediment*, https://doi.org/10.1007/978-3-030-15911-5_3

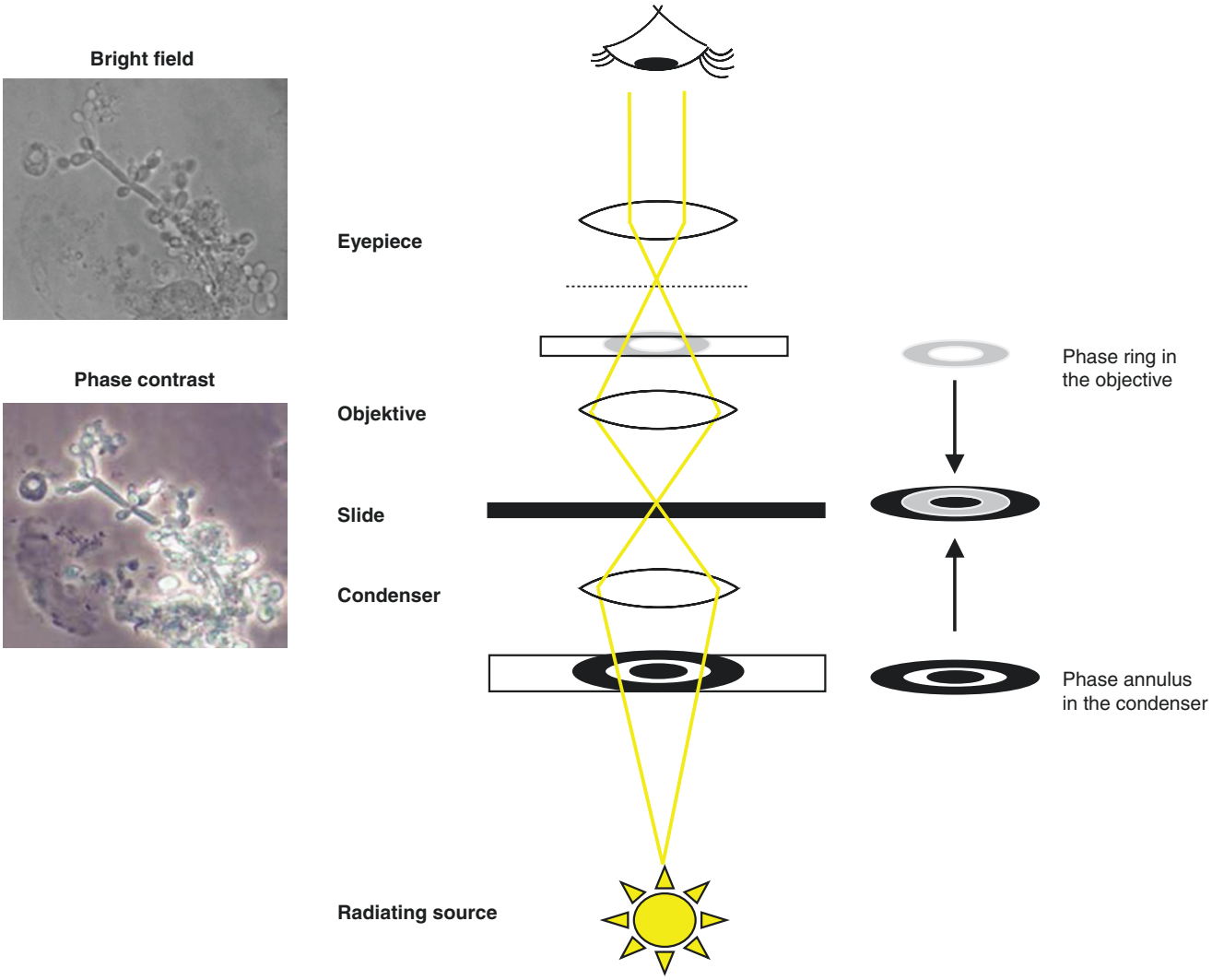


Fig. 3.1 The light pathway of phase-contrast microscopy

Fig. 3.2 PhaCo 40× objective with phase ring, marking: Ph 2

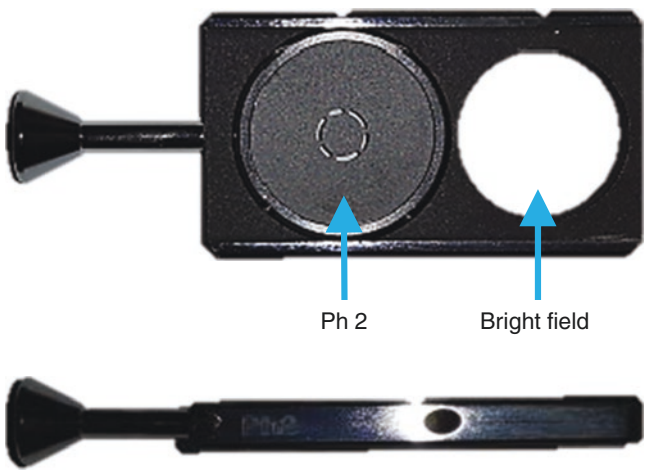


Fig. 3.3 Phase-contrast slider for the condenser with phase annulus Ph 2 and bright-field setting (top and side view)

Abbé Condenser

2

Phase annulus (Ph 1, Ph 2, Ph 3) for the corresponding lens or bright-field setting
E.g.: 40x- PhaCo-objektive = phase annulus 2 in condensor

1

Centering knob for the condenser phase annulus

3

Dial for adjusting the respective phase annulus or bright-field setting

Simple phase-contrast attachment for the Leitz condenser

Fig. 3.4 Zeiss Abbé condenser and simple phase-contrast attachment for the Leitz condenser

3.5 Centering the Phase Rings

Centering the phase annulus on the condenser is performed using the auxiliary microscope (Figs. 3.5 and 3.6). To achieve this, the auxiliary microscope is inserted in the observation tubes rather than the eyepiece (Fig. 3.6). Alternatively, a simple **diopter** can be used for centering rather than an auxiliary microscope.

In order to guarantee the phase contrast effect, centering needs to be performed and checked from time to time (as soon as phase-contrasting becomes weaker).

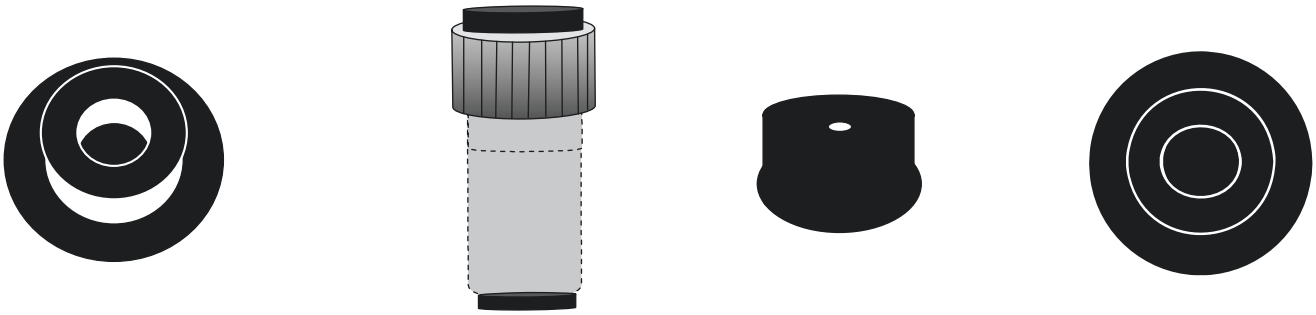


Fig. 3.5 Centering using an auxiliary microscope or diopter

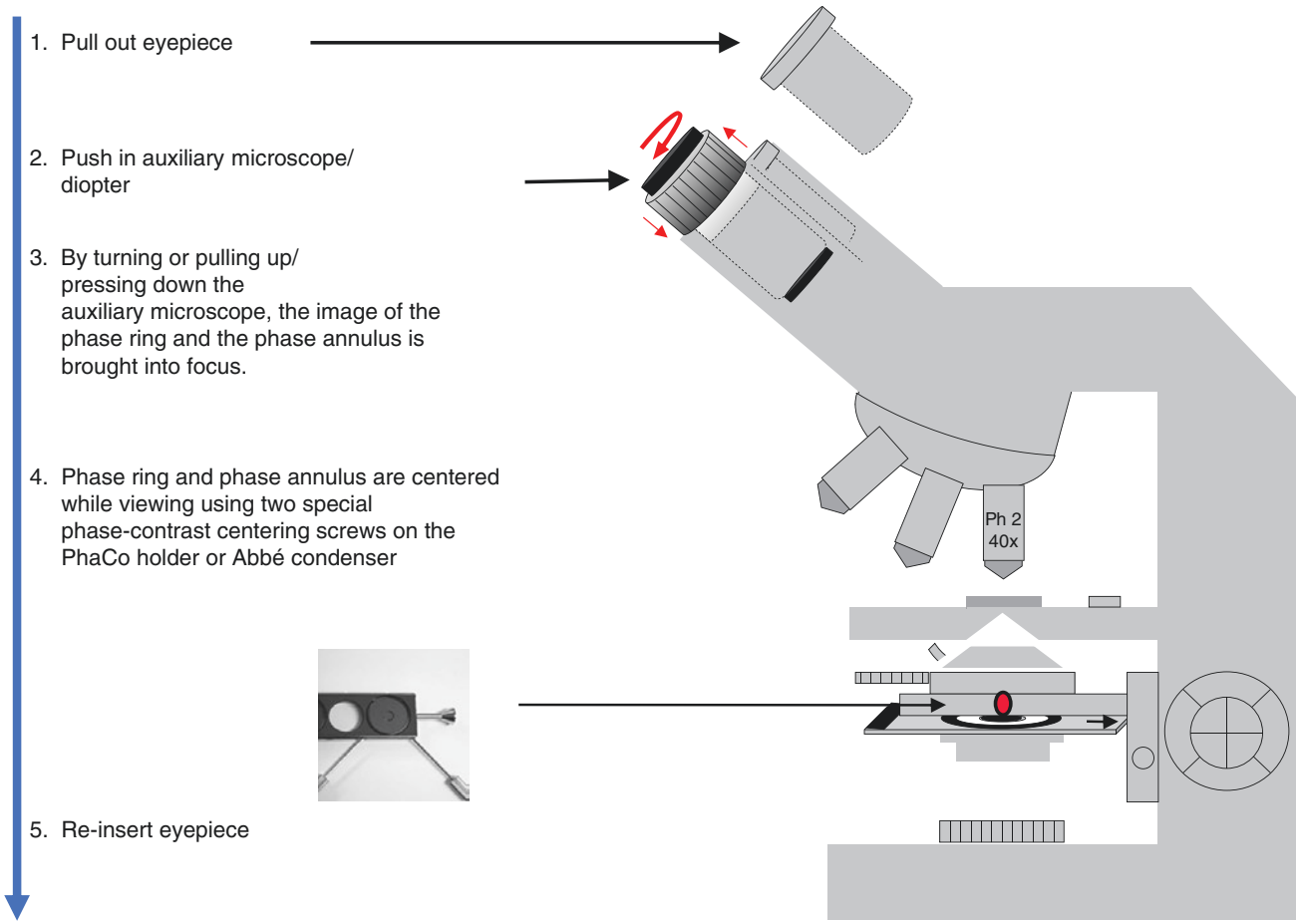


Fig. 3.6 Phase-contrast microscope: centering the phase rings

Macroscopic Urinalysis

4

4.1 Color

Depending on concentration, urine is light yellow to dark yellow in color. A noticeable deviation in color from the norm can indicate pathology or be harmless in nature.

4.1.1 Some Examples

- Colorless to light yellow
  Cause: polyuria, glycosuria in diabetes mellitus
- Dark yellow to orange
  Cause: oliguria, anuria, vitamin preparations
- Dark yellow to brownish-yellow
  Cause: hemoglobin and hemoglobin degradation products (bilirubin, porphyrins), drugs
- Milky/cloudy
  Cause: leukocyturia, salts, crystals
- Red to reddish-brown
  Cause: erythrocytes, myoglobin, urates, drugs, beetroot
- Dark brown to black
  Cause: erythrocytes, massive hemolysis

4.2 Odor

Certain foods, drugs, and bacteria alter the typical odor of urine.

4.2.1 Some Examples

- Extremely intensive odor
  Cause: garlic, asparagus
- Smells like chocolate, highly aromatic
  Cause: vitamin preparations, tropical fruits, spices
- Smells of ammonia
  Cause: urea-splitting bacteria
- Smells foul, putrid
  Cause: urinary tract infection
- Smells of fruit, acetone
  Cause: ketonuria

4.3 Cloudiness

Fresh urine at body temperature is normally clear. The colder and more concentrated a urine sample becomes, the more salts and crystals precipitate and cause turbidity or cloudiness. Urine also becomes visibly cloudy in the case of a pathological accumulation of bacteria or pyuria.

Only by analyzing solid components in urine (as in urinary sediment analysis) is it possible to conclusively identify the cause of cloudiness.

4.3.1 Some Examples

- Milky white
  Cause: bacteriuria, pyuria, phosphaturia, vaginal secretion
- Reddish (brick dust) upon cooling
  Cause: uraturia
- Red to reddish-brown
  Cause: macrohematuria
- Fat layer on the surface
  Cause: lipiduria in nephrotic syndrome, ointments, suppositories

J. Neuendorf, *Urine Sediment*, https://doi.org/10.1007/978-3-030-15911-5_4

Microscopic Urinalysis

5

5.1 Urine Sediment Preparation

Urine sample: fresh, midstream urine (preferably first urine in the morning), **not more than 2 h old**. Figure 5.1 shows the required equipment and materials.

5.1.1 Performance

- Mix urine well!
- Always pour the same amount of urine (**approximately 10 ml**) from the urine sample cup or urine vial into a **conical plastic sediment tube**.
- Apply urine to a test strip and analyze.
- To obtain the sediment, the urine sample is centrifuged for **8–10 min at 400 g** in a swing-out centrifuge, i.e., the centrifuge is set to 1620 U/min at a rotor radius of 13.2 cm, (see Sect. 5.4).
- The supernatant is decanted in one go without the sediment being agitated or poured out.
- **Carefully** resuspend the remaining sediment with the residual urine. (Do not shake the sediment excessively or tap it against the edge of the table to mix it up, since this can cause the casts to dissolve.)

5.2 Error Checklist and Tips for Urine Sediment Preparation

Although following the above-mentioned instructions for sediment preparation is straightforward, it is important to note that the actual performance of these steps needs to be carried out under exactly the same conditions. Only in this way can one obtain **reproducible results**.

The following small errors can have enormous consequences:

- The urine sample is kept for longer than 2 h at room temperature.
- The urine probe is not mixed. Urine samples are insufficiently mixed particularly when using urine sample containers without a lid.
- Arbitrary volume of urine taken for sediment preparation.
- Centrifuge time too long/too short.
- Centrifugal force altered, i.e., > or <400 g.
- Urine supernatant only slowly, i.e., not completely, decanted. One obtains an excessively large residual volume of urinary sediment (see Tip).
- Urine sediment not resuspended.

Tips

- Examine the sediment macroscopically for quantity and color here.
- If it is not possible to analyze the urine sediment immediately, store it in a closed place at 4–8 °C.
- If only a small urine sample of 4–6 ml is available, the small sample volume must be noted in the findings.
- The counting chamber method is particularly suitable for urine samples of only small volumes to determine the number of erythrocytes and leukocytes. The urine must not be centrifuged with this method. Native urine is used here (see Chap. 9).
- Hang out clear short instructions in the laboratory.
- Decanting: hold the test tube upside down and count to 3, then turn the test tube again and stand it upright.

J. Neuendorf, *Urine Sediment*, https://doi.org/10.1007/978-3-030-15911-5_5

Fig. 5.1 Equipment and materials

5.3 Discussion: Types of Centrifuge (Fig. 5.2)

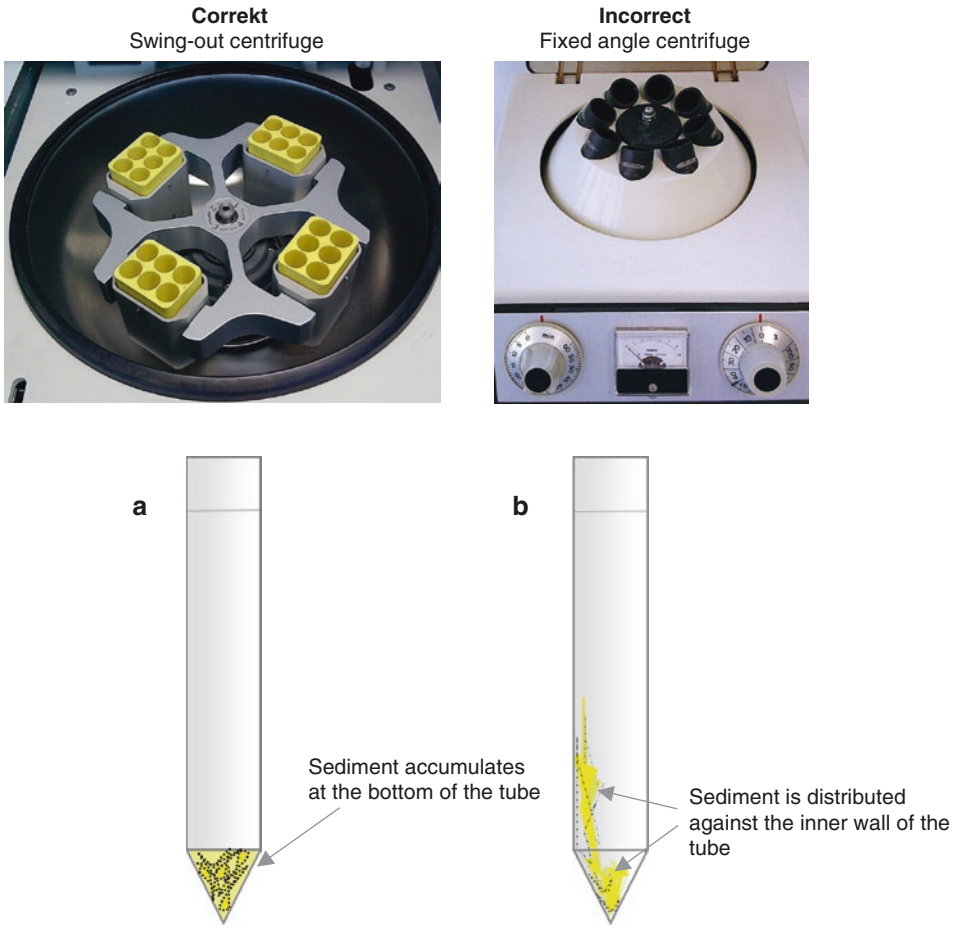


Fig. 5.2 Centrifuge types and centrifugation results. (**a**) If urine is centrifuged in a swing-out centrifuge, sediment forms in the lower conical part of the tube. (**b**) If a fixed-angle centrifuge is used, whereby sediment tubes cannot swing out, but instead are constantly tilted at a fixed angle, one obtains sediment that is drawn up against the wall in the lower part of the tube. This sediment is difficult to resuspend

5.4 Centrifuge Nomogram

To check that the centrifuge has the correct number of revolutions per minute, one measures the centrifuge rotation radius and e.g. uses the nomogram to check which number of revolutions per minute needs to be set at the relative centrifugal force (RCF) of 400 g (Fig. 5.3).

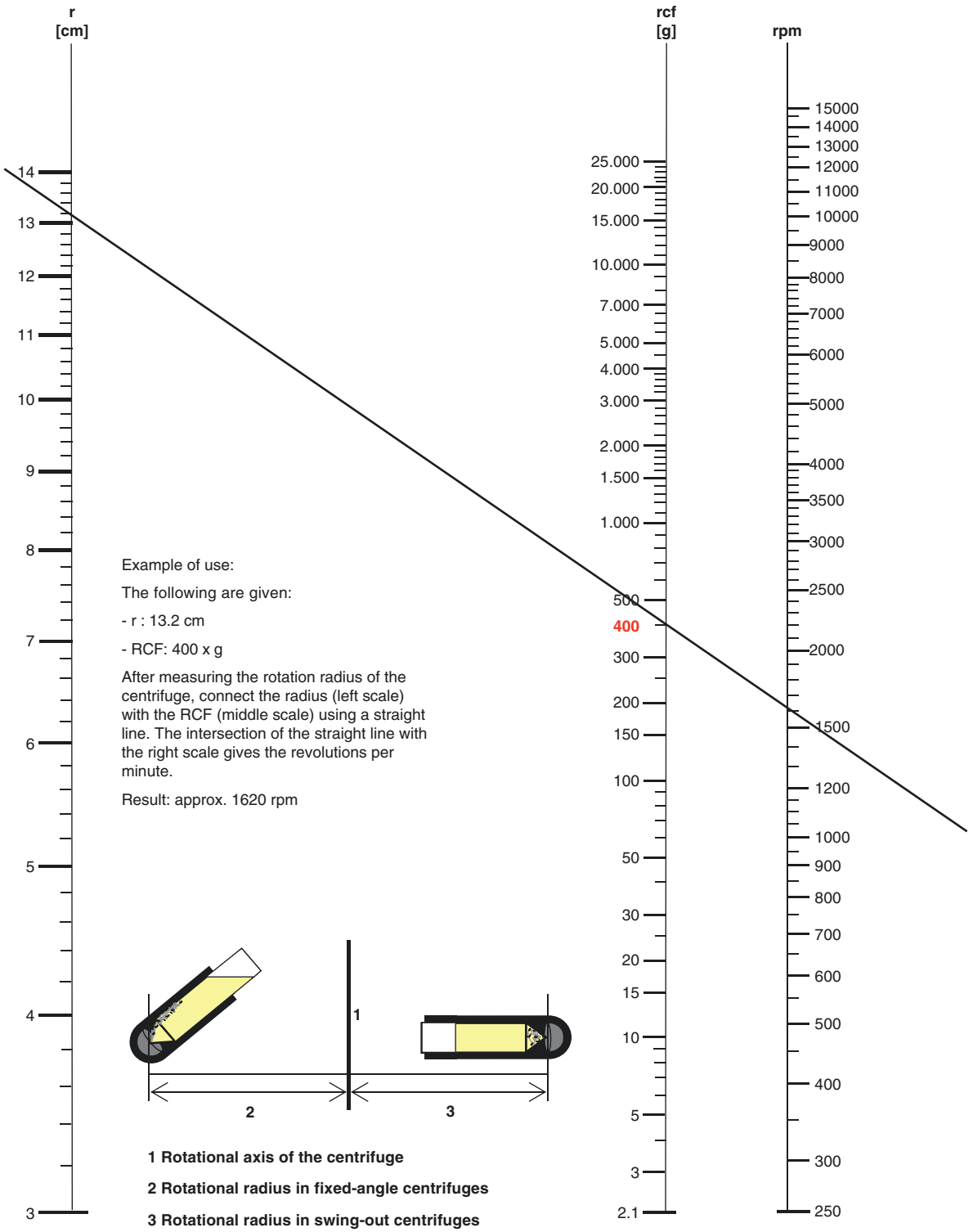


Fig. 5.3 Centrifuge nomogram. *r* Centrifugal radius, *RCF* relative centrifugal force, *rpm* revolutions per minute

5.5 Preparing the Native Sample

5.5.1 Materials (Fig. 5.4)

5.5.2 Performance

– Using a plastic dropping pipette, place a small drop (approximately 20 µl) of urine sediment on the center of a clean slide and cover with a clean cover glass (hold at edges), paying attention to avoid air bubbles (Fig. 5.5).
– To achieve this, one side of the cover glass is placed directly on the drop of urine sediment on the slide and slowly (using the pipette) lowered in such a way that the sediment beneath the cover glass is well distributed.
– The weight of the cover glass ensures that the sediment drop is distributed evenly (Fig. 5.6).
– The cover glass should not float away, nor should air bubbles be visible under the cover glass.

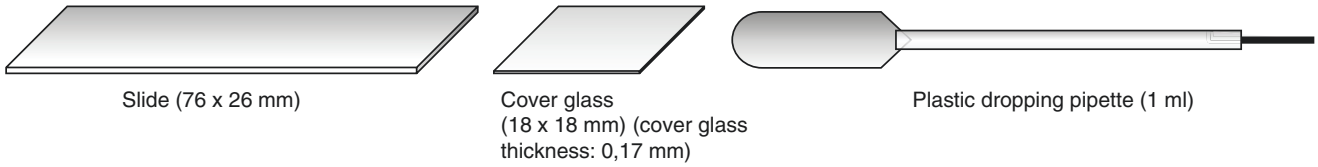


Fig. 5.4 Materials

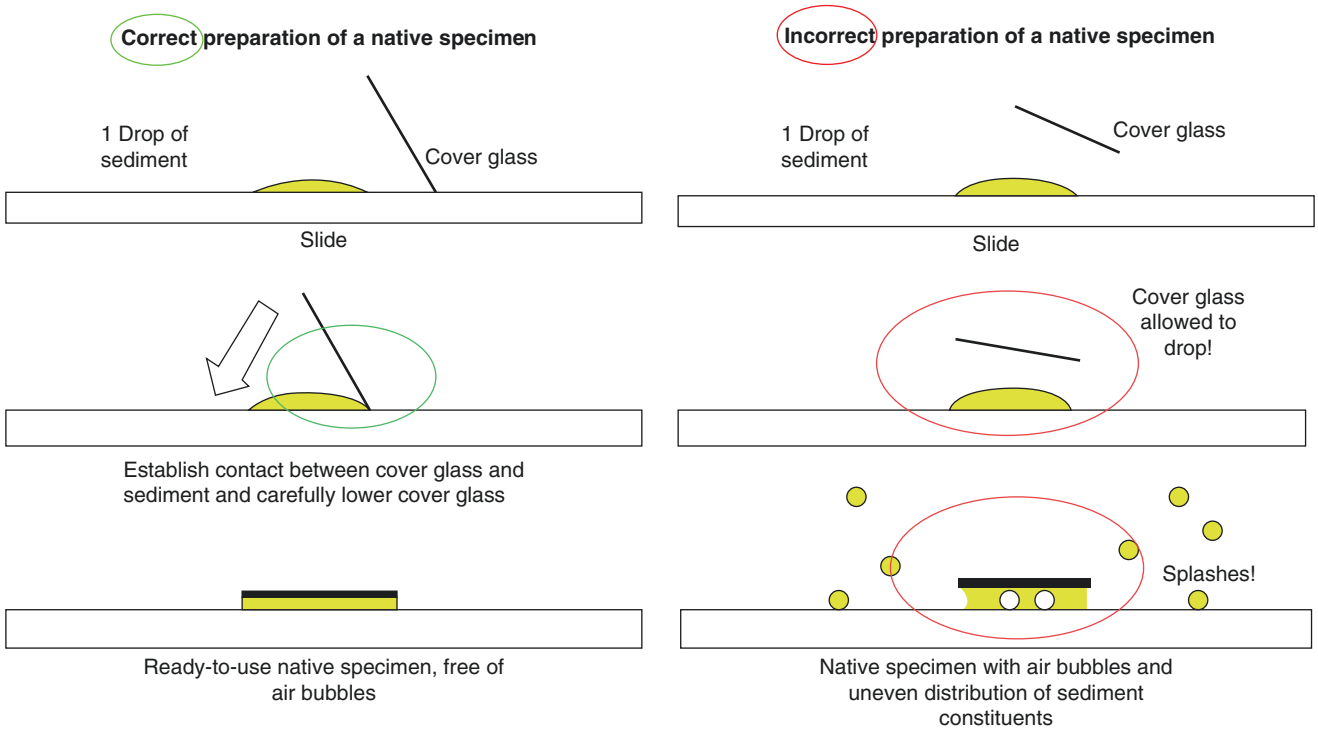


Fig. 5.5 Preparing a native specimen

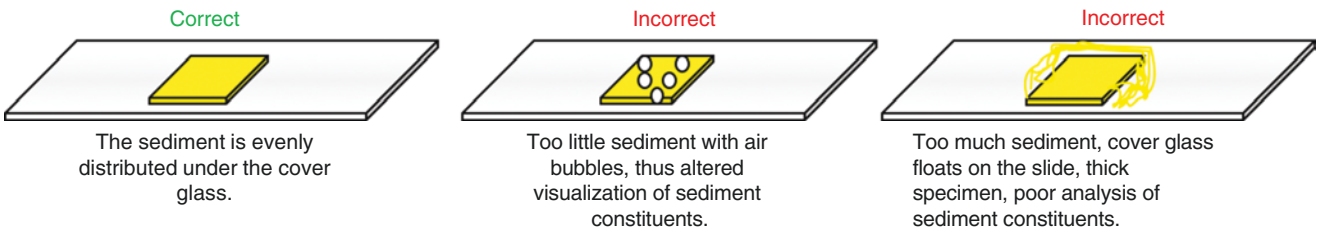


Fig. 5.6 Sediment distribution under the cover glass

– Since the finished native specimen dries rapidly, it must be examined immediately. If this is not possible, the finished specimen should be placed temporarily in a humidity chamber.

5.6 Switching the Microscope Between Bright-Field and Phase-Contrast

5.6.1 Switching the Microscope from Bright-Field to Phase-Contrast Microscopy

Phase-Contrast Microscope

– Place the PhaCo 40× objective (Ph 2) and the corresponding phase annulus on the condenser in the beam path, fold the front lens (if there is one) in or out according to the manufacturer's instructions, and open the aperture diaphragm and the field diaphragm, since phase contrast needs a lot of light (Fig. 5.7)!

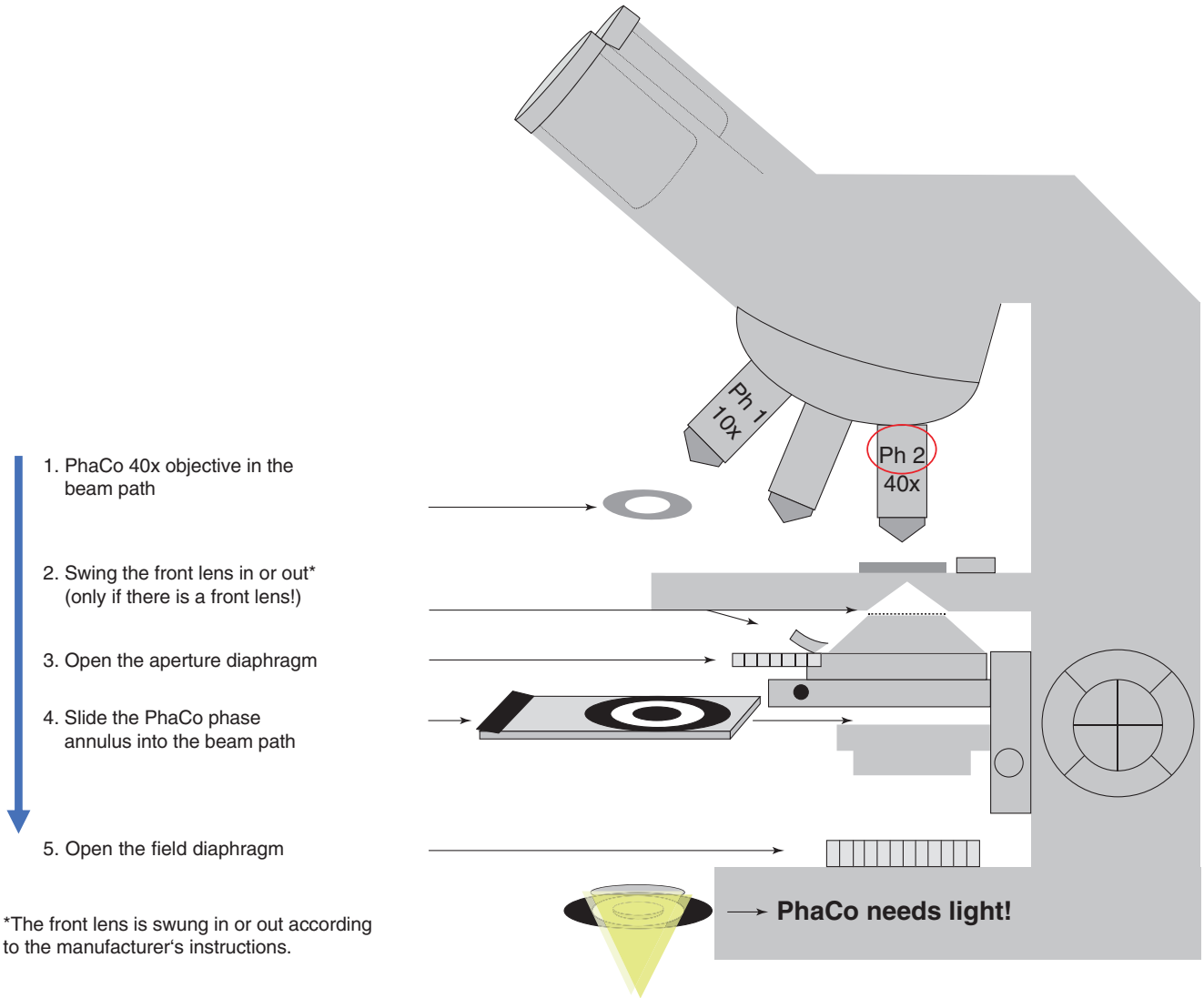


Fig. 5.7 Switching the microscope from bright-field to phase-contrast microscopy

5.6.2 Switching the Microscope from Phase-Contrast to Bright-Field Microscopy

Bright-Field Microscope

– PhaCo 40x objective is also used in bright-field (!); remove phase annulus on the condenser, close aperture diaphragm and field diaphragm slightly, front lens remains swung out (Fig. 5.8).

5.7 Specimen-Specific Adjustment of the Microscope

– In a first step, the **microscopic plane** of the specimen is set with the **10× objective**. Finding the correct plane can be challenging if the specimen is too thin or contains insufficient urinary sediment constituents. The plane can be seen more readily by adjusting the edge of the cover glass.
– The sediment is then analyzed for casts using a **10× objective**. Large urinary sediment constituents can often be found at the edge of the cover glass (Fig. 5.9).
– Following this, the sediment is analyzed per field of view using a **40× objective**.
– Irrespective of the magnification used, micrometer fine-focus adjustment must be constantly operated in order to fine-tune the microscopic plane to ensure that no components are overlooked.
– Microscopy analysis is carried out primarily in phase-contrast mode.
– Bright-field is used to evaluate colors (crystals, salts) and clusters of constituents/cells.

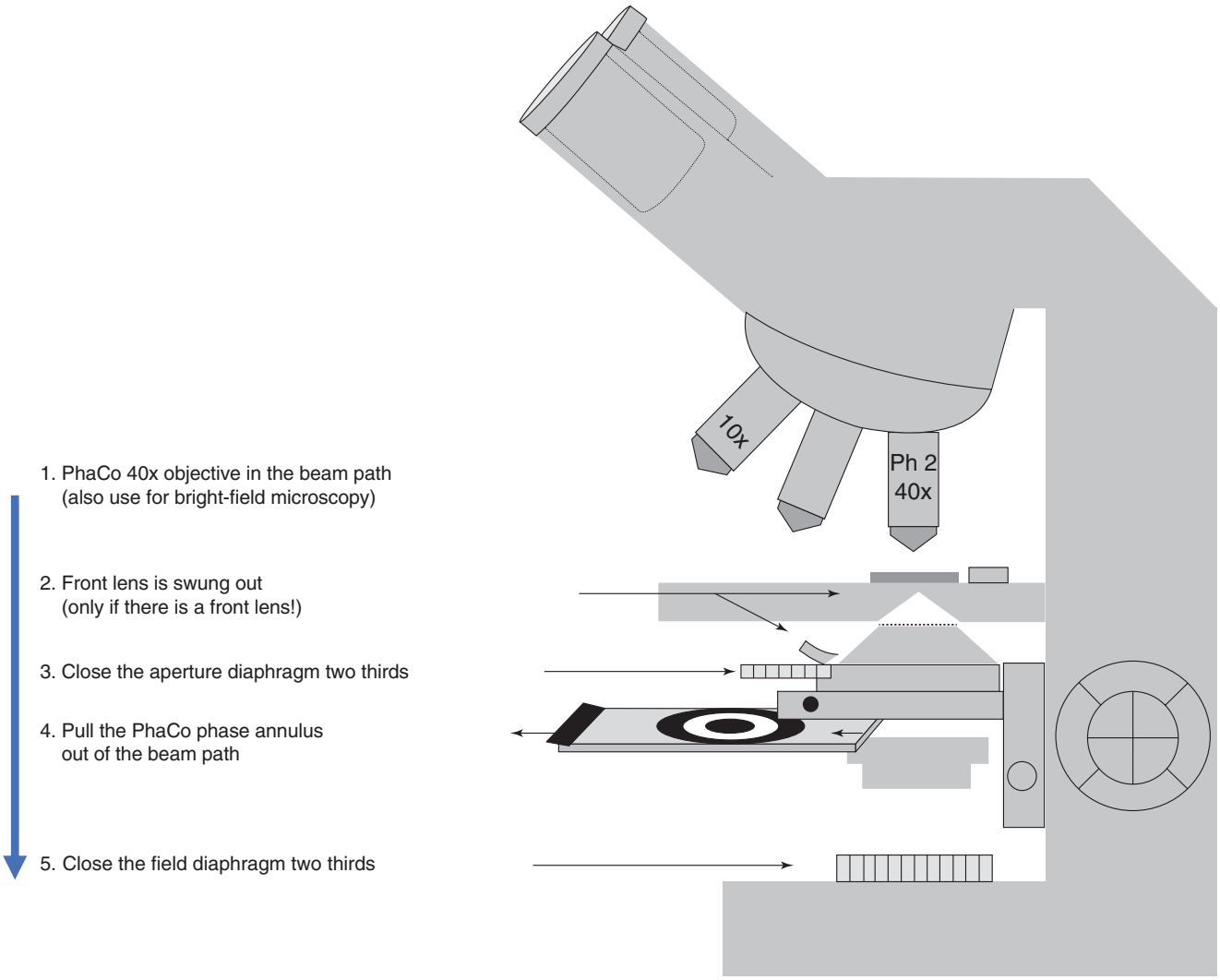


Fig. 5.8 Switching the microscope from phase-contrast to bright-field microscopy

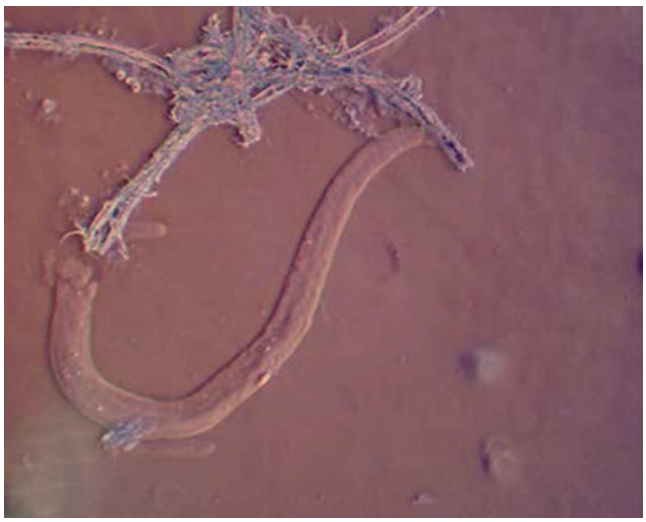


Fig. 5.9 10× objective, one large and two smaller casts, artifacts

5.8 Semi-quantitative Analysis/Units

A semi-quantitative evaluation is carried out by examining 20–30 high power fields (HPF) in a meandering pattern at 400× magnification (corresponding to an 10× eyepiece and a 40× objective) while considering a certain field number (Fig. 5.11).

The following classifications are available for organized sediment constituents, i.e., **erythrocytes**, **leukocytes**, and **epithelial cells**, whereby the minimum and maximum number per HPF are given (Fig. 5.10).

Casts are generally rarer than organized sediment constituents and are difficult to identify using these classifications (Fig. 5.10). Therefore, one should add the respective cast type when looking through the HPF and the total of all HPF (aHPF) evaluated should be given as the result.

Sediment constituents such as **bacteria**, **yeast cells**, **crystals**, **salts**, and **spermatozoa** are not counted, but instead given as **crosses** (Fig. 5.12).

Unit

- High power field = **HPF** (400x magnification)
- **Special feature**: To indicate casts, one can use **all** HPF evaluated as a unit = **aHPF**

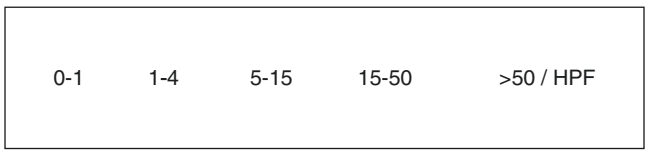


Fig. 5.10 Classification of organized sediment components

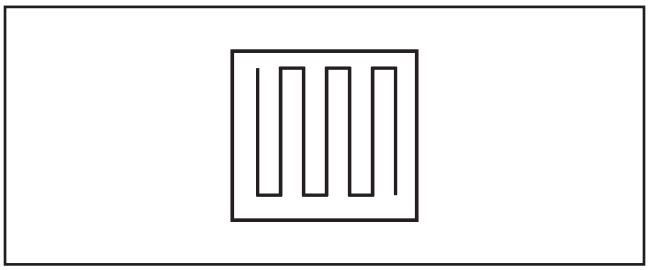


Fig. 5.11 Examination in a meandering pattern

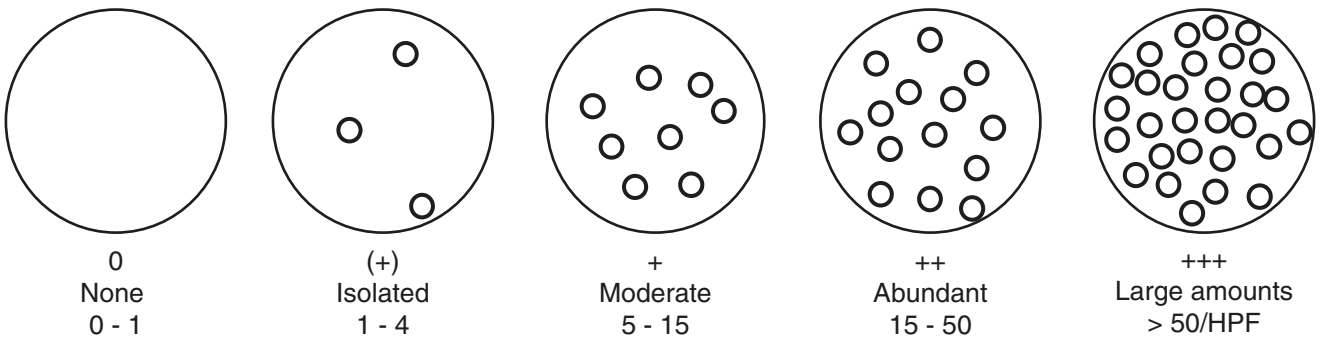


Fig. 5.12 Sediment constituents shown with crosses

5.9 Discussion: Field Number and Normal Values

The normal values of the individual urinary sediment constituents must be evaluated on the basis of the eyepiece/field number used and the objective or the size of the resulting HPF in order to better compare and interpret them internally and externally (Fig. 5.13). The size of the HPF depends on the eyepiece/field number used and the objective. In addition to the magnification, the eyepiece also shows the field number, which gives the diameter of the field diaphragm in millimeters.

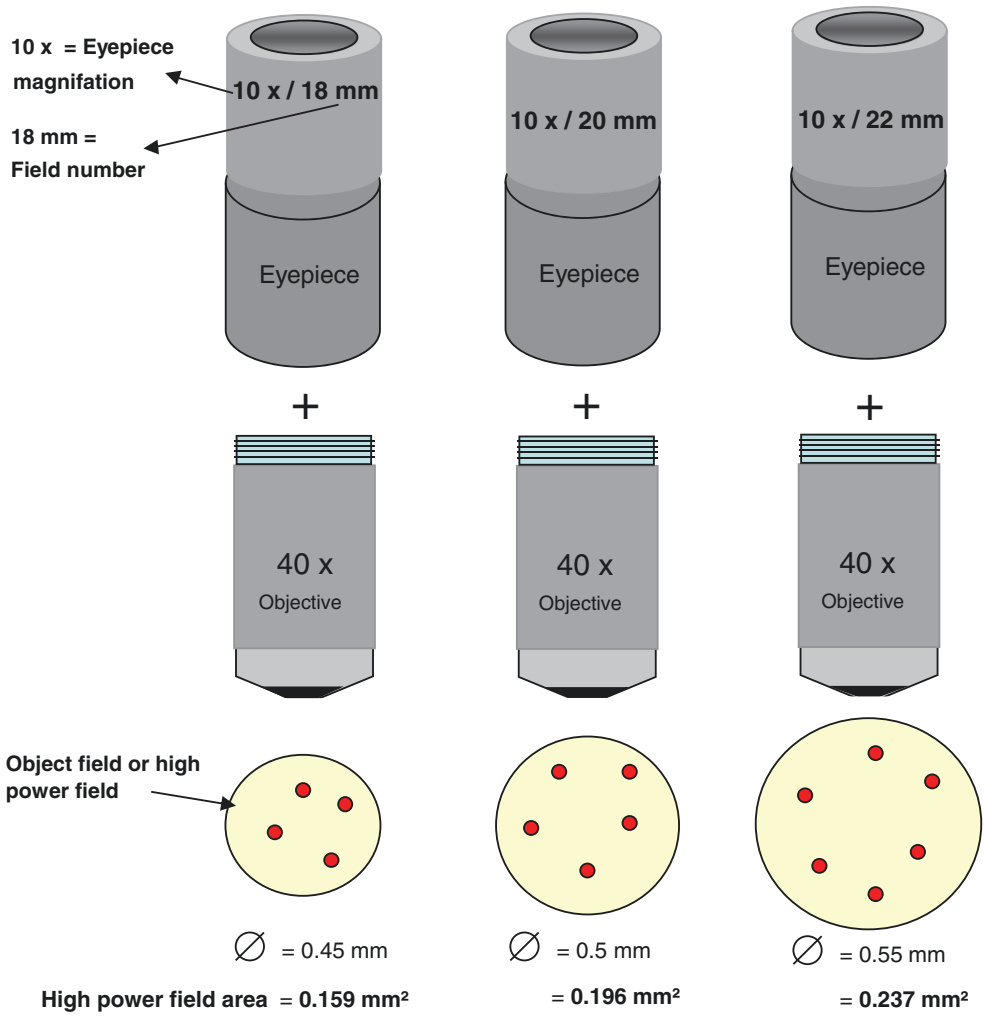


Fig. 5.13 Field number and normal values

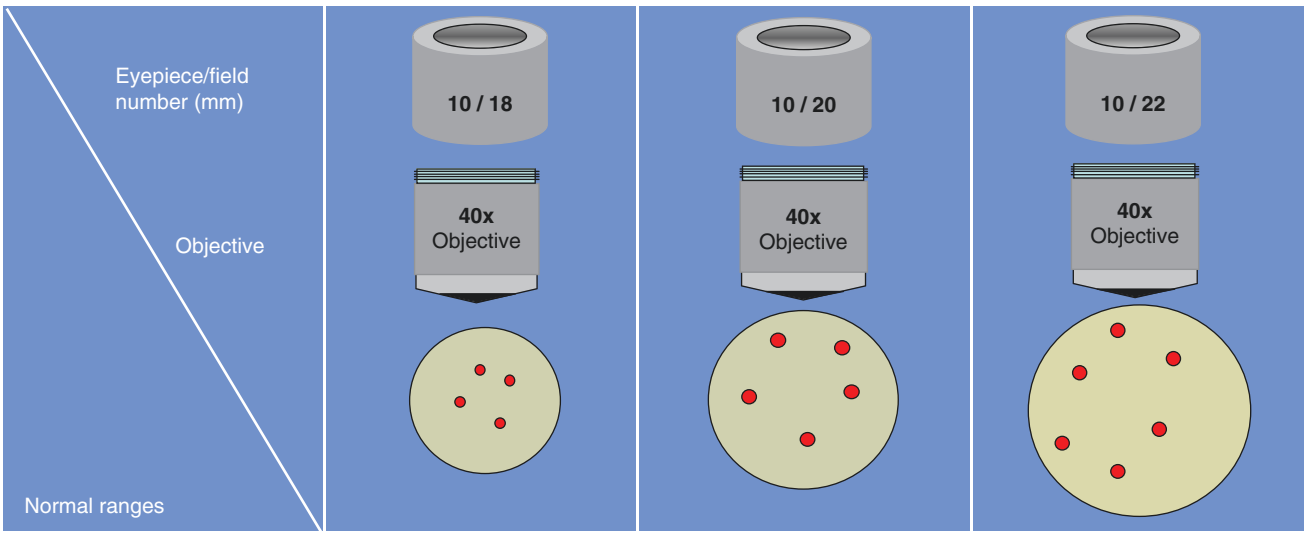
– Example: 10×/18, i.e., 10× eyepiece and field number = 18 mm

The larger an eyepiece's field number, the larger the area in the specimen that can be microscopically visualized, the so-called object field or HPF. The sizes of the object field/HPF are calculated as follows:

– Field number/Scale denominator of the objective = diameter of the object field/HPF
– $\pi \cdot r^2$ = Object field/HPF (area) (mm^2)

The normal values on which this book is based (Chap. 7) relate to 400× magnification and a field number of 18 mm (Fig. 5.14).

> Conclusion: If four cells per HPF are counted with a small field number (e.g., 18 mm), six cells per visual field correspond to a large field number (e.g., 22 mm).



EumEc	0–1	0–1	0–2
Lc	1–4	1–5	1–6
SqEc	0–15	0–19	0–22
TransEc	0–1	0–1	0– 2

Fig. 5.14 Normal value ranges corresponding to the respective field number and the corresponding HPF size

Anatomy of the Kidneys and Urinary Tract System

6

Figure 6.1 shows the anatomy of the kidneys and urinary tract system.

The urine constituents found in urine sediment are named according to their origin. From a diagnostic perspective, it is important to distinguish cells of renal origin from cells of post-renal origin. For example, the increased presence of dysmorphic erythrocytes and acanthocytes is attributed to impaired renal tissue or impaired glomeruli. Eumorphic erythrocytes are more likely to be of post-renal origin (exception: renal tumor). Casts form in the distal renal tubules depending on the pH value and concentration of the urine.

Epithelial cells are also named according to their origin. Renal and tubular epithelial cells are found in kidney tissue. Transitional epithelium/urothelium covers the renal pelvis and urinary tract system up to the upper segment of the urethra. The squamous epithelium originates in the lower segment of the urethra.

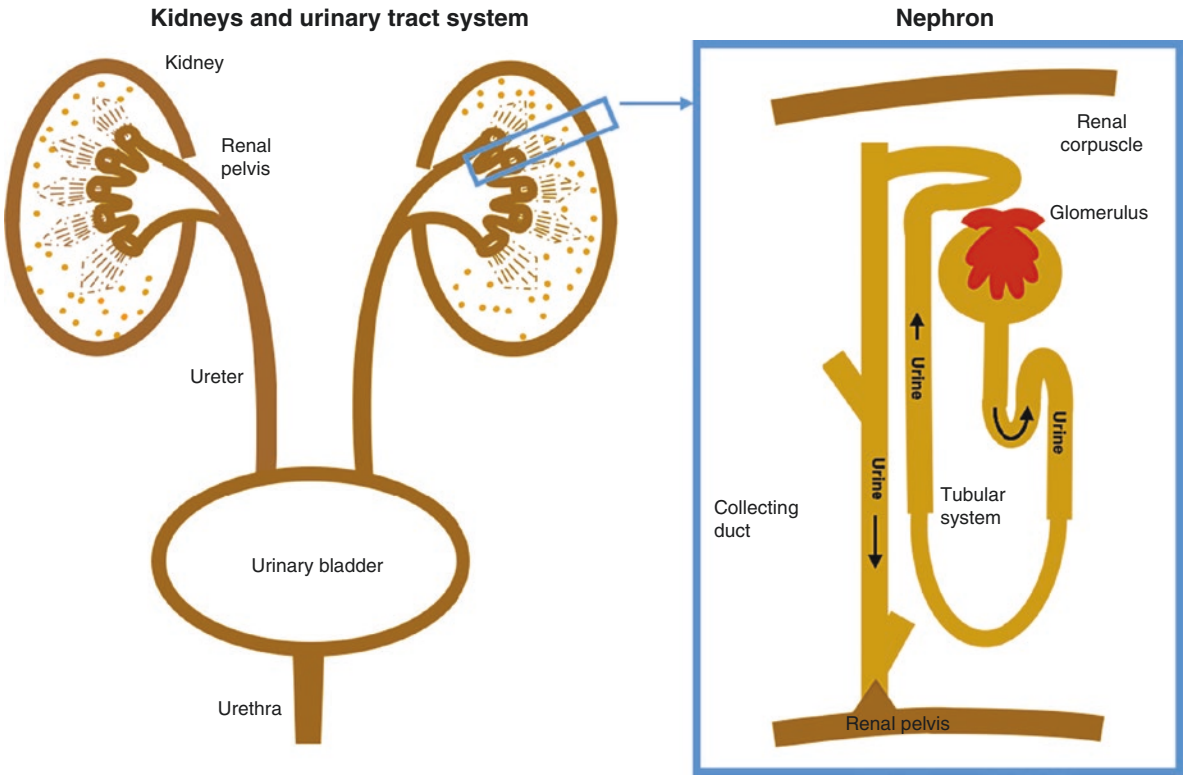


Fig. 6.1 Overview: anatomy of the kidneys, urinary tract system, and nephron

J. Neuendorf, *Urine Sediment*, https://doi.org/10.1007/978-3-030-15911-5_6

Description of Urinary Sediment Constituents

7

Figure 7.1a, b provides an overview—and permits a comparison—of urinary sediment constituents in photographic and schematic representations.

7.1 Erythrocytes

7.1.1 Hematuria (Increased Excretion of Erythrocytes in Urine)

- Macrohematuria: Urine assumes a red color due to massive excretion of erythrocytes.
- Microhematuria: Urine is yellow-colored. Erythrocytes can only be detected chemically (with the urine test strip) and microscopically.

7.1.2 Eumorphic Erythrocytes—NR: 0–1/HPF

7.1.2.1 Biconcave, Disc-Shaped Erythrocytes (pH = 6)

Morphology: Round cells, clear yellowish-red color in bright field, contain no nucleus or granules, smaller than leukocytes (Fig. 7.2).

Due to the erythrocyte's biconcave shape, one sometimes sees a double contour. The eumorphic erythrocyte changes its shape according to the pH value of the urine:

7.1.2.2 Thorn Apple-Shaped Erythrocytes (pH < 6)

In hypertonic urine, the erythrocyte loses water, becomes smaller, and takes on the typical thorn-apple shape. The surface of the erythrocyte has an abundance of small, pointed projections (Fig. 7.3).

7.1.2.3 Erythrocyte Ghosts (pH > 6)

In hypotonic urine, the erythrocyte absorbs water, loses as a result its biconcave shape, and enlarges to a light disc. The contour of the "erythrocyte ghost" is sometimes only poorly visible (Fig. 7.4).

Disorder: Hematuria in kidney tumors, urinary tract tumors, stones, urinary tract infections, trauma.

7.1.3 Dysmorphic Erythrocytes

Morphology: Predominantly microcytic, highly diverse, distinct from eumorphic erythrocytes. In addition to numerous morphological changes not described here, one sees the following typical changes: ring-shaped, wavy ring-shaped, slitted ring shape, fragmentocytes, pin, or sphere internally and externally (for classification see Thiel 1986) (Fig. 7.5).

Also in the case of non-glomerular haematuria, it is assumed that up to 20% of erythrocytes can exhibit dysmorphic characteristics [S. Roth, *Urinzytologie und Sedimentanalyse*, Springer 2018 (5th edition), p. 171]:

- <20% Dysmorphic erythrocytes: no renal disease.
- 20–70% Dysmorphic erythrocytes: equivocal suspicion of renal disease.
- >70% Dysmorphic erythrocytes: suspected kidney disease.

Dysmorphic erythrocytes can be identified particularly well in phase-contrast mode. The acanthocyte is a special form of dysmorphic erythrocyte.

- **Disorder**: kidney disease.

J. Neuendorf, *Urine Sediment*, https://doi.org/10.1007/978-3-030-15911-5_7

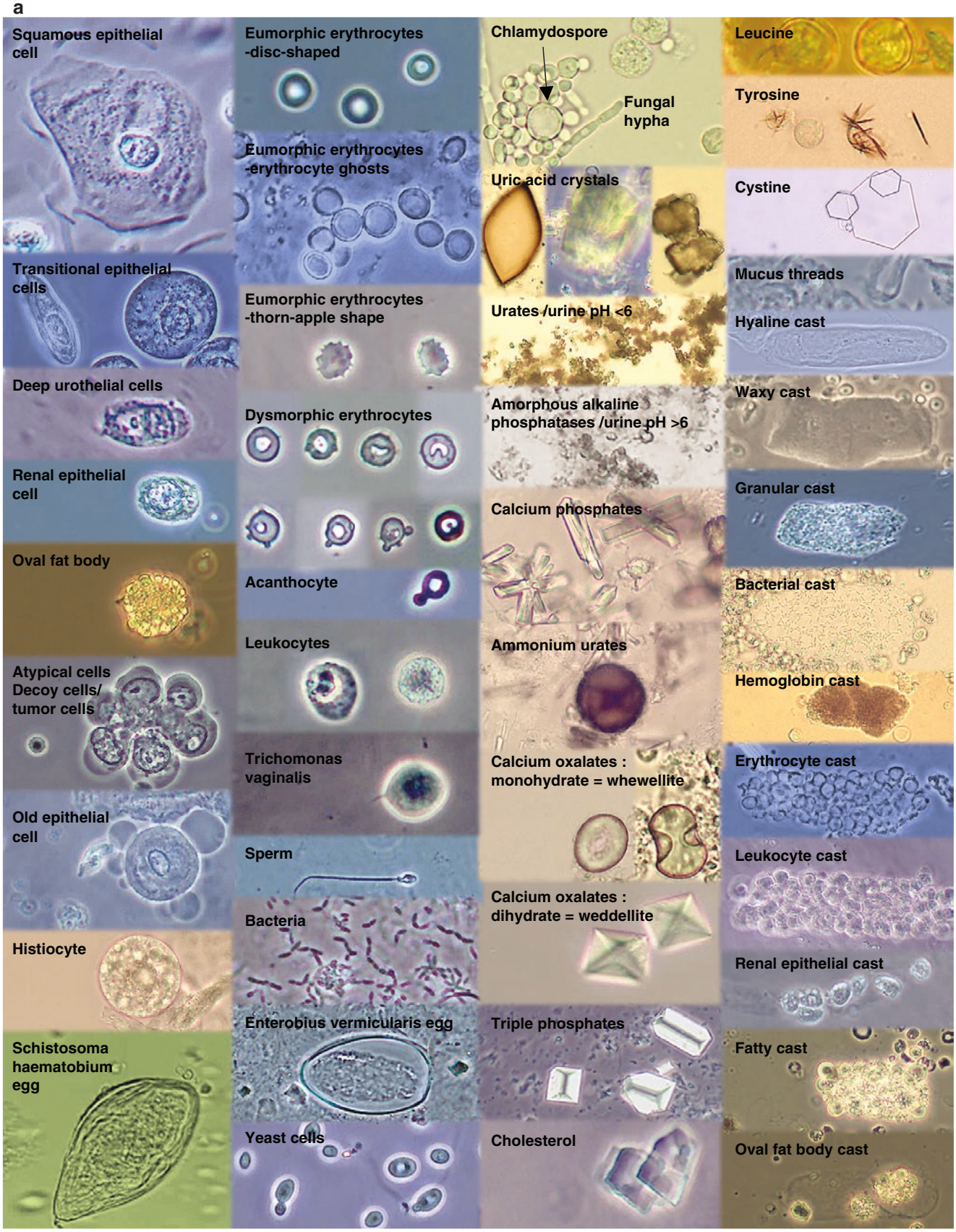


Fig. 7.1a Urinary sediment constituents. (**a**) Photographic representation

b

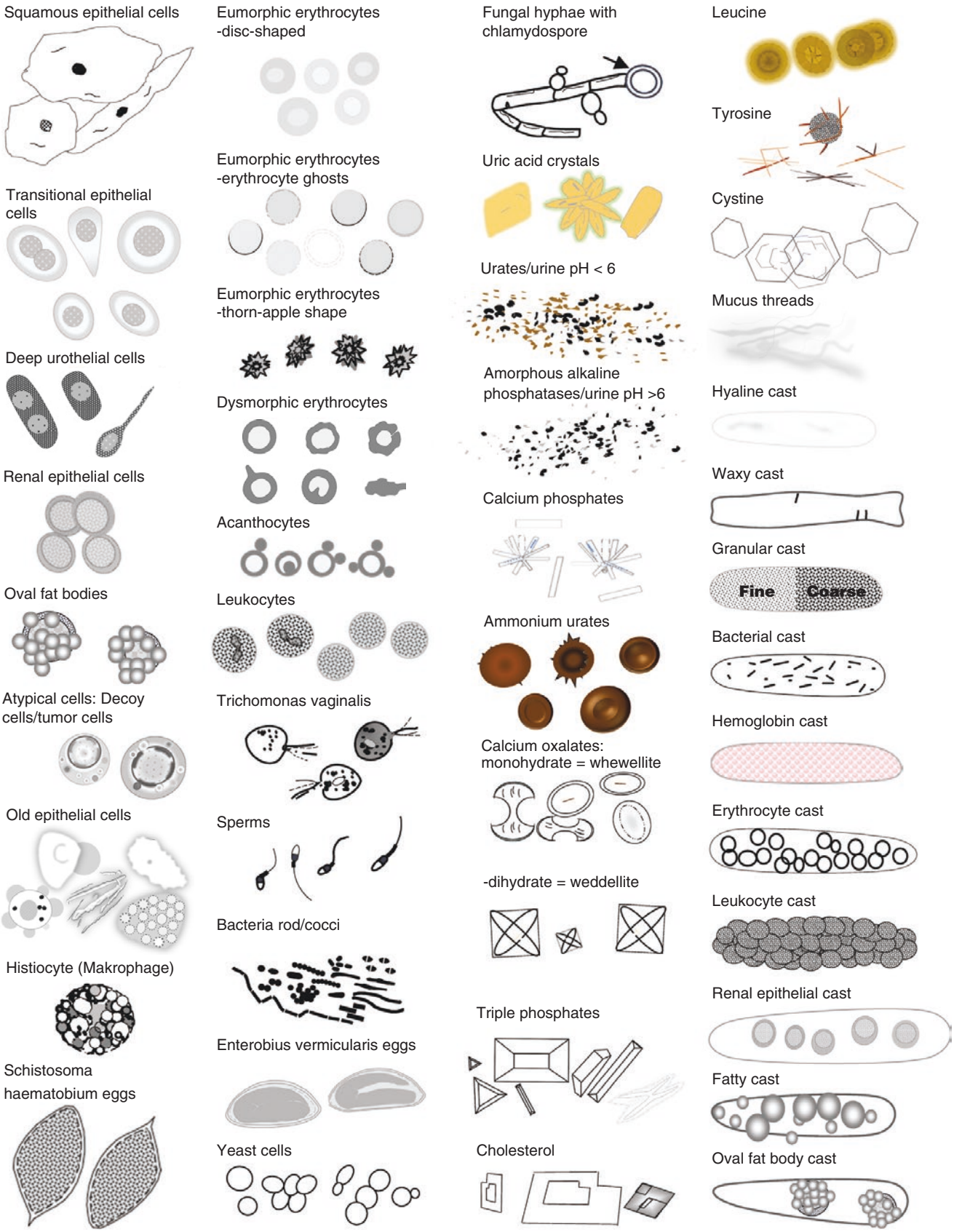


Fig. 7.1b (continued) Schematic representation

Fig. 7.2 Biconcave, disc-shaped erythrocytes

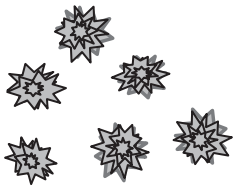


Fig. 7.3 Thorn apple-shaped erythrocytes

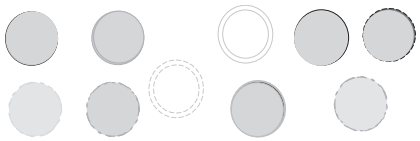


Fig. 7.4 Erythrocyte ghosts

Fig. 7.5 Dysmorphic erythrocytes

7.1.4 Acanthocytes—NR: <5%

Morphology: The acanthocyte has at least one spherical projection, while the thorn apple has regular, smaller, and more pointed projections. The projections on the acanthocyte appear as exo-and endospheres. These typical spheres form during passage through the damaged glomeruli (Fig. 7.6).

– **Disorder**: glomerulonephritis.

7.2 Leukocytes

7.2.1 Leukocytes—NR: 1–4/HPF

Morphology of the segmented granulocytes and classification into two cell types. Small leukocyte: round cell, somewhat larger than a biconcave erythrocyte, with a dark and granular surface. Large leukocyte with visible, segmented nucleus and granules in the cytoplasm. In old urine, the leukocytes swell and the nucleus is clearly visible. The cells lie individually and in clusters (Fig. 7.7). Leukocyturia: increased excretion of leukocytes (> 4 /HPF at 400× magnification, field number 18 mm). Pyuria: Massively increased excretion of leukocytes, urine is cloudy white, and the sediment is whitish and viscose.

Fig. 7.6 Acanthocytes

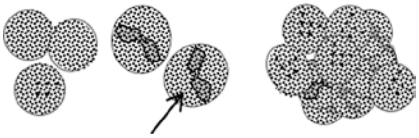


Fig. 7.7 Leukocytes, small and large type, clusters. *Arrow*, typical nuclear structure of a large, segmented granulocyte

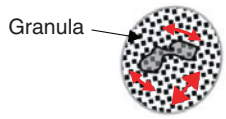


Fig. 7.8 Sternheimer-Malbin cell = leukocyte (greatly enlarged here) red arrows indicate mobile granules in the cytoplasm

– **Disorder**: Inflammation in the urinary tract system, inflammatory renal disease.

Note Leukocytes such as lymphocytes may be excreted in urine in the case of renal transplant rejection or eosinophilic granulocytes in the case of allergic reactions. Lymphocytes cannot be detected using the leukocyte test area on the urine test strip.

7.2.2 Special Forms of Leukocytes

7.2.2.1 Sternheimer-Malbin Cells or Bright Cells

Morphology: In some leukocytes (segmented granulocytes), one sees a characteristic granule motility (Fig. 7.8).

Note: This characteristic granule motility is clearly visible in the short video (see Chap. 11, Sect. 11.7.5)

7.2.3 Histiocytes (Macrophages)—NR: None

Morphology: Larger than granulocytes, vary greatly in size, contain granulation and abundant vacuoles that appear as large, bright and dark circles. Nucleus: variform and only hazily visible. These cells belong to the monocyte–macrophage system. They are able to phagocytose bacteria or cells such as erythrocytes. They are easily mistaken for round epithelial cells or oval fat bodies (Fig. 7.9).

– **Disorder**: inflammatory process

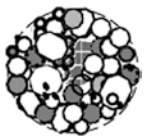


Fig. 7.9 Histiocyte

7.3 Epithelial Cells

Note If urine remains in the catheter bag for a prolonged period of time, the appearance of epithelial cells may diverge from the norm. They should not be mistaken for tumor cells!

7.3.1 Squamous Epithelial Cells—NR: 0–15/HPF

Morphology: Largest epithelium in urine, polygonal border. Nucleus: small, compact, dark, and opaque to slightly transparent with a perinuclear area of brightening, i.e., perinuclear halo (phase contrast), occur as single cells and in clusters (Fig. 7.10).

In old urine, the outer border of the squamous epithelial cell becomes increasingly round. An increased presence of squamous epithelial cells in urine can be an indication that the urine specimen was not a midstream sample.

– **Origin**: Squamous epithelial cells are found in the lower urinary tract and the external genital area. The urine sediment of females frequently contains an increased number of squamous epithelial cells that originated from vaginal secretion.

Note An increased presence of squamous epithelial cells in urine can produce a false positive result on the leukocyte test area on the urine test strip. Therefore, urinary sediment analysis should always be additionally performed in order to confirm the diagnosis.

7.3.2 Transitional Epithelial Cells or Urothelial Cells—NR: 0–1/HPF

Morphology: Size varies greatly, predominantly round to oval, but sometimes also with a tail, opaque cytoplasm. Nucleus: large, round, loose chromatin, sometimes also two nuclei. The cells lie separately and/or in clusters. Small transitional epithelial cells cannot always be unequivocally distinguished from renal epithelial cells (Fig. 7.11).

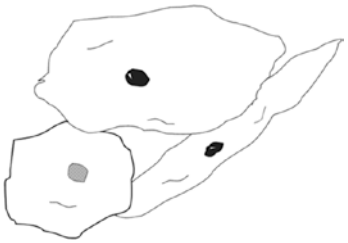


Fig. 7.10 Squamous epithelial cells

Fig. 7.11 Transitional epithelial cells. *Arrow*, transitional epithelial cell with tail

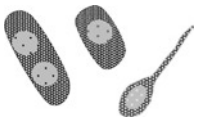


Fig. 7.12 Deep urothelial cells

– **Origin**: urinary tract system, i.e., renal pelvis, ureter, bladder, urethra.
– **Disorder**: Inflammation of the urinary tract system. If atypical cells and cell groups are found, further urine cytology testing should be performed. In the unstained native specimen, tumor cells cannot be reliably identified or unequivocally differentiated from virus-infected cells.

7.3.3 Deep Urothelial Cells—NR: None

Morphology: Epithelium with cubic and tailed configuration, smaller than transitional epithelium, dark, fine-grained cytoplasm, partially with granules. Nucleus: one or two bright, transparent cell nuclei. These epithelial cells need to be differentiated from renal epithelial cells (Fig. 7.12).

– **Origin**: The urothelium is a multi-layered epithelium. In the case of chronic urinary tract infections, the upper epithelial layer of the transitional epithelial cells erodes. Further epithelial layers, as well as deep urothelial cells, are found below and can be seen in urine sediment.

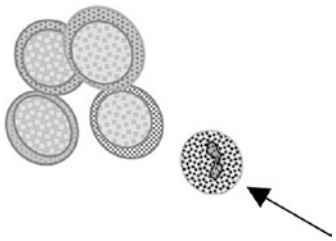


Fig. 7.13 Renal or tubular epithelial cells. *Arrow*, leukocyte for comparison

– **Disorder**: Injury to the urothelium, chronic urinary tract infection.

7.3.4 Renal or Tubular Epithelial Cells—NR: None

Morphology: Smallest epithelial cell in urine, slightly larger than a leukocyte, dark, fine-grained cytoplasm, narrow cytoplasmic rim, sometimes with granules. Nucleus: homogeneous vesicular nucleus with a bright structure, cells lie separately or in clusters (Fig. 7.13).

– **Origin**: Kidneys
– **Disorder**: tubular damage, toxic damage, and transplant rejection

7.3.5 Oval Fat Bodies—NR: None

Morphology: Tubular epithelial cells with fat droplets. The round- to oval-shaped cells can become enormous depending on how many lipids are stored. Cell contour and cell nucleus are sometimes no longer visible. The fat droplets are extremely bright and shiny (Fig. 7.14).

– **Origin**: Kidneys
– **Disorder**: Nephrotic syndrome

7.3.6 Virus-Infected Cells

7.3.6.1 Example: Decoy Cells—NR: None

The tubular/urothelial cell infected with polyomavirus is called a decoy cell. It is called a decoy cell since it is easily mistaken for an atypical cell.

Morphology: Epithelium with enlarged, ground glass-like nucleus, chromatin is unevenly distributed and lies condensed on the nuclear membrane (Fig. 7.15).

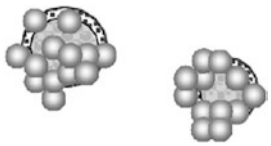


Fig. 7.14 Oval fat bodies

Fig. 7.15 Decoy cells

– **Origin**: Renal tissue and urothelium from the renal pelvis to the ureter
– **Disorder**: Polyomavirus infection in the setting of weakened immune system, important in kidney transplantation

Note Decoy cells are difficult to differentiate from old leukocytes/renal epithelial cells in older urine samples.

7.3.7 Discussion: Cell Description

7.3.7.1 Criteria

It is sometimes highly challenging to identify and differentiate small epithelial cells. In the case of difficulties classifying cells or when the cells appear atypical, one can describe the cell. As part of this, the following criteria should be described:

– Position: single/in a cluster (Fig. 7.16)
– Anisocytosis (size differences) of the cells (Fig. 7.17)
– Nucleus–cytoplasm ratio (Fig. 7.18)
– Nuclear shape/nucleoli/chromatin (Fig. 7.19)
– Cell size/nucleus size (Fig. 7.20)
– Inclusions: Vacuoles, granules, other (Fig. 7.21)

7.3.8 Discussion: Morphological Criteria of Old Cells and Epithelial Cells

A decaying, aging cell (such as leukocytes and epithelial cells) may exhibit a single morphological characteristic or several different characteristics (Fig. 7.22). Typical characteristics include:

– The nucleus can no longer be clearly visualized.
– The boundaries between cell nucleus and cytoplasm are indistinct.

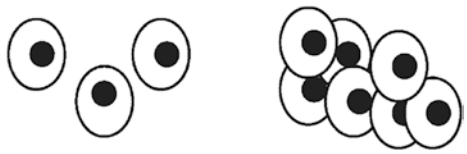


Fig. 7.16 Position: single/in a cluster

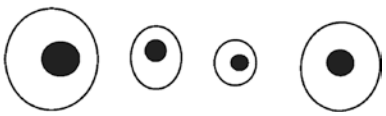


Fig. 7.17 Cell anisocytosis

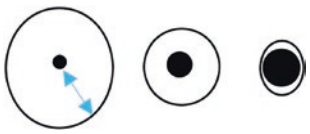


Fig. 7.18 Nucleus–cytoplasm ratio

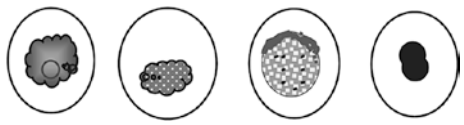


Fig. 7.19 Nuclear shape/nucleoli/chromatin

Fig. 7.20 Cell size/nucleus size

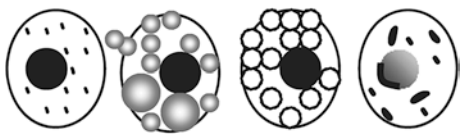


Fig. 7.21 Inclusions: vacuoles, granules, other

Fig. 7.22 Morphological criteria of old cells

– The cytoplasm has a wrinkled appearance.
– The cytoplasm may be permeated by one or more vacuoles.
– The cell contour is irregular and has a frayed appearance.
– The cell contour appears to be swollen.
– One or more aging-related bubbles surround the outer cell edge and/or overlay the cell.

7.4 Casts

Morphology: Cylindrical structures with sharp contours and rounded ends in highly varying widths and lengths.

Casts are made up of a large-molecule protein, the Tamm–Horsfall protein, or uromodulin. This glycoprotein is released into the kidneys by tubular cells in the tubules. Depending on the concentration and pH value of the urine, it aggregates and precipitates. The casts form in different widths according to the diameter of the tubule (Fig. 7.23).

If casts are made up only of the Tamm–Horsfall protein, they are referred to as hyaline casts. It is possible for cells such as erythrocytes, leukocytes, epithelial cells, as well as fat particles, etc., to be stored and to accumulate in the protein substance. Casts dissolve rapidly in alkaline urine.

Casts develop in the distal renal tubule and are suggestive of kidney disease. Exceptions here include hyaline and granular casts, which are of no differential diagnostic relevance. They can be detected in cases of fever, severe generalized disease, and renal disease, as well as in healthy individuals.

If proteinuria is detected in a urine sample, particular attention should be paid to cylinduria in microscopic urinary sediment analysis. The specimen is analyzed for casts using a 10× objective. Cast type is identified with a 40× objective.

The following cast types are shown here:

– Hyaline casts
– Granular casts
– Waxy casts
– Renal Epithelial casts
– Erythrocyte casts
– Leukocyte casts
– Fatty or lipid casts and oval fat bodies casts
– Hemoglobin casts and myoglobin casts
– Bacterial casts
– Pseudocasts or mucus threads

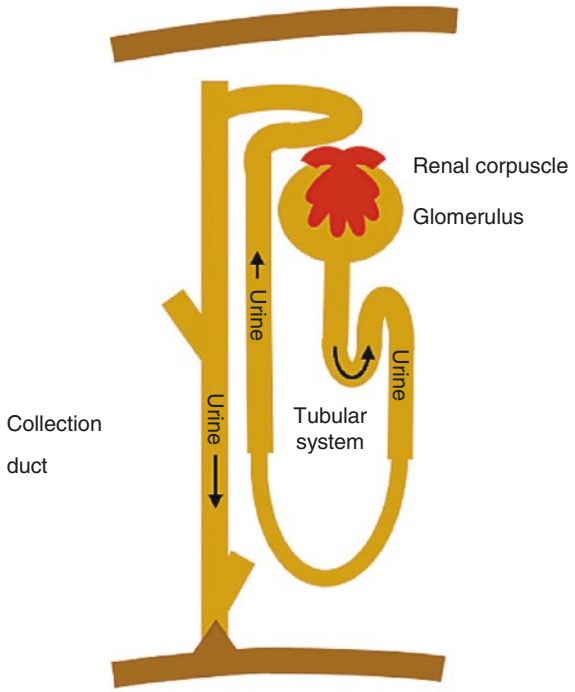
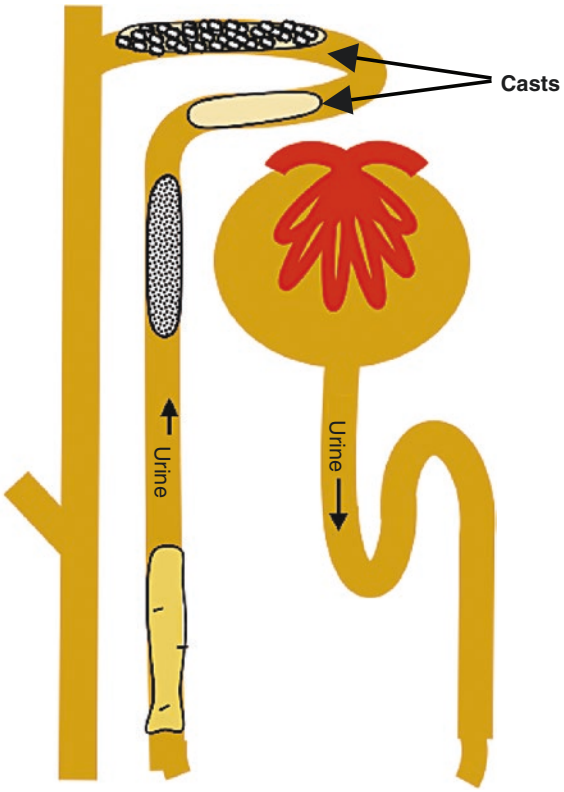


Fig. 7.23 Nephron. Detail: distal tubule with casts

Fig. 7.24 Hyaline cast



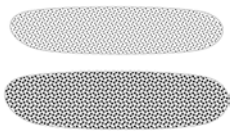


Fig. 7.25 Granular casts

7.4.1 Hyaline Casts—NR: Isolated

Morphology: Colorless, matt, transparent, isolated small dots/threads, otherwise empty, different lengths and thicknesses. In order to avoid confusing these with mucus threads, attention is paid to whether the lateral edges are continuous and whether both ends of the cast are rounded and not frayed (Fig. 7.24).

Since they are highly transparent, hyaline casts are easily overlooked in bright-field mode. However, they are readily identified in phase-contrast mode.

– **Disorder**: Fever, kidney disease, physical exertion

7.4.2 Granular Casts—NR: None

Morphology: Fine or coarse, non-crystalline granules, cast size varies considerably, granules are not necessarily distributed over the entire cast. The granules are composed of plasma proteins (Fig. 7.25).

Note Cylindrical accumulations of amorphous crystals should not be confused with granular casts!

– **Disorder**: severe generalized disease, renal disease

7.4.3 Waxy Casts—NR: None

Morphology: Refractive, transparent, wax-like. Pronounced, somewhat angular ends and lateral indentations are typical. Waxy casts are composed of denatured plasma proteins. In bright-field mode, the contour of the waxy cast appears as if drawn with a pencil and, unlike the hyaline cast, cannot be overlooked in bright-field mode (Fig. 7.26).

– **Disorder**: severe renal disease

Fig. 7.26 Waxy cast

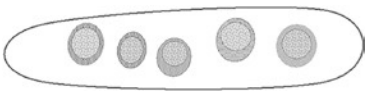


Fig. 7.27 Renal Epithelial cast

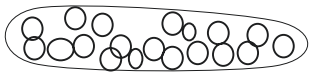


Fig. 7.28 Erythrocyte cast

7.4.4 Renal Epithelial Casts—NR: None

Morphology: Embedded renal tubular epithelial cells are extremely difficult to identify.

It is not easy to distinguish an epithelial cast from a leukocyte cast. Therefore, the European Urinalysis Guidelines recommend only referring generally to a cell cast in the case of doubt.

The nucleus of the epithelium appears more rounded and somewhat lighter than the nucleus of a granulocyte, which is darker and segmented (Fig. 7.27).

– **Disorder**: acute kidney failure, tubular damage

7.4.5 Erythrocyte Casts—NR: None

Morphology: Colorless to yellowish-brown, some contain abundant, some only scant erythrocytes. Small, transparent, round cells are readily identified. Cast size varies considerably (Fig. 7.28). Their presence suggests renal hematuria.

– **Disorder**: interstitial nephritis, glomerulonephritis

7.4.6 Leukocyte Casts—NR: None

Morphology: Dark, gritty cells, the size of casts and the quantity of leukocytes vary considerably; the abundant leukocytes obscure the contour of the cast.

> Important to remember: Whereas cells in a leukocyte cast appear smaller due to the high packing density, the leukocytes loosely assembled in a cluster appear larger.

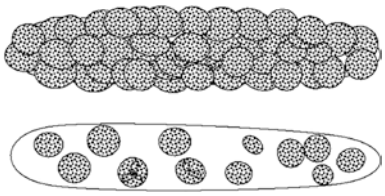


Fig. 7.29 Leukocyte casts

Fig. 7.30 Fatty or lipid cast and oval fat bodies cast

Fig. 7.31 Hemoglobin cast and myoglobin cast

Whereas small, transparent, round cells are typical of erythrocyte casts, one can identify leukocyte casts from the somewhat larger cells with their dark, gritty surface (Fig. 7.29).

– **Disorder**: pyelonephritis

7.4.7 Fatty or Lipid Casts and Oval Fat Bodies Casts—NR: None

Morphology of fatty casts: Filled with variously sized and highly luminescent fat droplets. Fat can be readily and rapidly stained with Sudan IV for better identification.

Morphology of oval fat bodies casts: These contain oval fat bodies, i.e., round tubular epithelial cells packed with fat. The more embedded the fat is, the more difficult it is to identify the epithelial cell (Fig. 7.30).

– **Disorder**: nephrotic syndrome

7.4.8 Hemoglobin Casts and Myoglobin Casts—NR: None

Morphology: Yellowish to brownish-red hemoglobin casts that are granular in appearance. Distinguishing between these two types of cast is challenging. They are found in hemoglobinuria and myoglobinuria (Fig. 7.31).

Note In the case of muscle injury, serum creatinine kinase is elevated.

Fig. 7.32 Bacterial cast

Fig. 7.33 Mucus threads (pseudocasts)

– **Disorder**: hemolysis, severe muscle injury

7.4.9 Bacterial Casts—NR: None

Morphology: Bacteria embedded in the cast, readily identifiable in phase contrast (Fig. 7.32).

– **Disorder**: severe bacterial urinary tract infection with renal involvement (pyelonephritis)

7.4.10 Mucus Threads (Pseudocasts)

Morphology: Wide and/or narrow, disordered fibrous structures without continuous contours at their ends (in contrast to casts). Cells, bacteria, and crystals often become entangled in the mucus threads (Fig. 7.33), which are made up of Tamm–Horsfall protein. Mucus threads are not recorded in findings.

Note Pseudocasts need to be distinguished from hyaline casts.

7.5 Microorganisms

7.5.1 Bacteria—NR: (+) - +/HPF

Morphology: Round and elongated.

As shown in Fig. 7.34, bacteria are extremely small and diverse. They can also be identified in the native specimen by their partial intrinsic motility and by their relatively rounded and non-luminescent contour. Thus bacteria can be distinguished from small, luminescent amorphous salts.

Bacteria occur individually, in chains, and/or in clusters.

Urine sediment findings do not take bacteria shape into account, but only quantity! Pathogen determination is the task of microbiology.

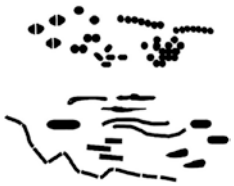


Fig. 7.34 Round bacteria (cocci) and elongated bacteria (bacilli, rod-shaped)

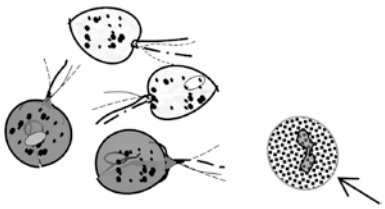


Fig. 7.35 Trichomonads. *Arrow*, leukocyte for comparison

If urine is left to stand, bacteria multiply rapidly: the bacterial count doubles in 20 min! If a urine sample is to be left standing for more than 2 h prior to analysis, it should be kept in a refrigerator.

– **Disorder**: urinary tract infection

7.5.2 Trichomonads (Flagellates)—NR: None

Morphology: Roundish-oval to pear-shaped with four flagellates, somewhat larger than leukocytes.

Trichomonads are identified microscopically in fresh urine by the fact that they rapidly migrate through the HPF. Phase-contrast microscopy clearly shows the flagellates that enable them to move. In contrast to a segmented leukocyte, the internal appearance of the trichomonad is more homogeneous. The cytoplasm may contain numerous granules (Fig. 7.35).

In the case of degeneration, trichomonads appear to be more rounded and may exhibit vacuoles. They can only be reliably analyzed in fresh urine that is still warm, since they stop moving upon cooling down, making them extremely difficult to distinguish from leukocytes or transitional epithelial cells.

– **Disorder**: trichomonad infection with the parasite Trichomonas vaginalis; this infection belongs to the group of sexually transmitted diseases.

7.5.3 *Schistosoma haematobium* Eggs—NR: None

Morphology eggs: At a length of 140–150 µm and a width of 40–70 µm, these are extremely large eggs with a typical termi-

nal spine (Fig. 7.36). The large Schistosoma eggs can be very well seen microscopically at a magnification as low as 100x.

The excretion of Schistosoma eggs can simultaneously result in leukocyturia and eumorphic hematuria and increased excretion of transitional epithelial cells. In order to reliably detect the eggs, the use of endstream urine rather than midstream urine is recommended. Urine samples should be collected between 10:00 AM and 14:00 PM, since this is generally the time of highest egg excretion. Urine should be protected from light in order to prevent the larvae from hatching.

– **Disorder**: pathogens responsible for bladder or urogenital schistosomiasis, trematode infection. Schistosomiasis is a tropical disease, i.e., it spreads primarily in inland waters in tropical and subtropical regions. Contact with inland waters bears the risk of infection.

7.5.4 *Enterobius vermicularis* (formerly *Oxyuris vermicularis*) Eggs—NR: None

Morphology: 20–30 µm wide and up to 50–60 µm long; with double-layered shell. Eggs are oval and colorless with one typically flattened side. Larvae are sometimes readily visible (Fig. 7.37).

– **Disorder**: pinworm infestation (*Enterobiasis* or *Oxyuriasis*). Pinworm infestation is one of the most widespread parasitic diseases. The eggs enter the urine due to contamination or poor hygiene.

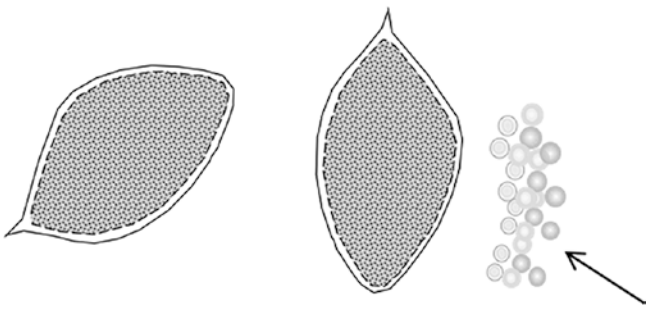


Fig. 7.36 *Schistosoma haematobium* eggs. *Arrow*, eumorphic erythrocytes for comparison

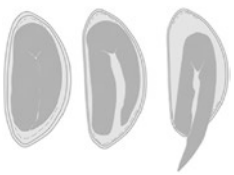


Fig. 7.37 *Enterobius vermicularis* eggs at varying stages of development

7.5.5 Yeasts—NR: None

Morphology of **yeast cells**: In bright-field mode, yeast cells appear colorless, while in phase-contrast mode they are light in color. They may be round, oval, or elongated. Yeast cells have a nucleus, which can sometimes be visualized at 400x magnification. Yeast cells of different sizes are often found together in pairs, exhibiting the typical "mother–daughter asymmetry." They are generally somewhat smaller than erythrocytes. Distinguishing between yeast cells and erythrocytes is challenging (Fig. 7.38).

Morphology of **fungal hyphae**: These are several adjacent tubular structures (pseudohyphae) that form a longer pseudomycelium with the typical transverse septa. They can become extremely long (100 µm), branch out, and form a network. Old fungal hyphae lose this tubular structure and cannot be differentiated from thicker mucus threads or filament-shaped bacteria (Fig. 7.38).

Morphology of **chlamydospores**: Chlamydospores are rarely found in urine sediment. These are survival structures that are only formed when the nutrient supply for the yeast is not optimal. Chlamydospores grow on a pseudomycelium, but may lie at some distance from the fungal hypha in a native specimen. Chlamydospores are somewhat larger than erythrocyte ghosts (risk of confusion!) (Fig. 7.39).

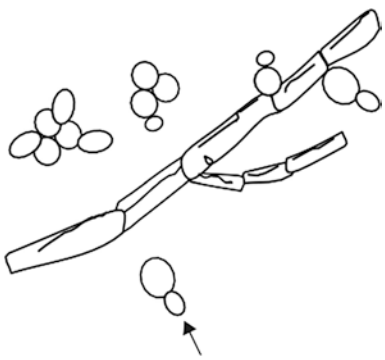


Fig. 7.38 Yeast cells and fungal hyphae. *Arrow*, "mother–daughter asymmetry"

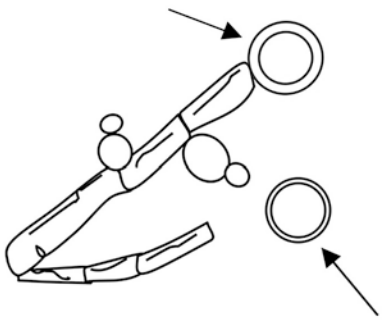


Fig. 7.39 Yeast hypha with yeast cells and chlamydospore (*upper arrow*); erythrocyte ghost for comparison (*lower arrow*)

– **Disorder**: Fungal infection, immunocompromised individuals, frequent in diabetics and following antibiotic treatment

7.6 Crystals

Crystals precipitate in urine in larger or smaller quantities depending on concentration, pH value, and temperature. Crystalluria is more likely to be detected in a morning urine sample than in a spontaneous urine sample. The various crystals can have highly diverse shapes and are readily identified microscopically. There are a number of pathological crystals that need to be reliably differentiated from non-pathological crystals. Some crystals are formed through the excretion of drugs in urine.

Crystalluria can be an indication of urolithiasis. The commonest urinary stones include:

– Calcium oxalate stones (whewellite and weddellite)
– Calcium phosphate stones (apatite/brushite)
– Magnesium ammonium phosphate stones (triple phosphates cause infection stones/struvite)
– Uric acid stones (uricite)

The urinary stones listed here often occur in combination.

Rarer types of urinary stone include, e.g.,:

– Cystine stones
– 2,8-Dihydroxyadenine stones
– Xanthine stones

In addition to crystalluria, other urine findings such as pH value, protein, nitrite, specific gravity, leukocytes, and erythrocytes are important in the diagnosis of urolithiasis.

Only the most important crystals are discussed in the following sections.

Cystine, leucine, tyrosine, and cholesterol are considered pathological crystals and point to severe liver damage or protein metabolism disorders. Cystine is of particular relevance, since cystinuria is indicative of a genetic reabsorption disorder in relation to various amino acids. These crystals are comparatively rare.

7.6.1 Cystine—NR: None

Morphology: Colorless, hexagonal shape, sometimes with irregular sides, thin discs, single and sometimes superimposed. In older urine samples, cystine crystals may take on a brownish color and the typical hexagonal contour is only partially or no longer visible. The identification of these crystals is of considerable clinical relevance and is indicative of cystinuria. Acidic urine pH (Fig. 7.40).

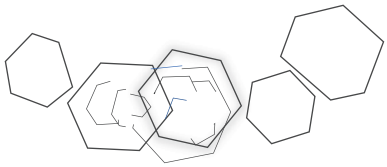


Fig. 7.40 Cystine

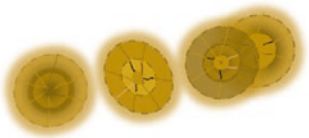


Fig. 7.41 Leucine

Note Only fresh urine samples should be used, since the morphologically altered cystine crystals in older urine samples can resemble other round, brown crystals, such as ammonium urate.

– **Disorder**: cystinuria (hereditary metabolic disease involving a tubular reabsorption disorder for cystine, arginine, ornithine, and lysine—only the amino acid cystine crystallizes at an acidic pH value), cystine stones

7.6.2 Leucine—NR: None

Morphology: Brownish-yellow in color, spherical with radial striation, acidic urine pH (Fig. 7.41).

Note Leucine is extremely rare and should not be confused with ammonium urate!

– **Disorder**: severe liver damage

7.6.3 Tyrosine—NR: None

Morphology: Extremely rare, found in acidic urine, shiny, colorless to yellow-brown, extremely fine needles, single or lying together (needle bundles), some of which are also found intracellularly in leukocytes. Leukocytes pierced by needles are particularly easy to identify in bright-field mode. In addition to tyrosine excretion, bilirubin deposits (granules or short needles) that light up yellow can be differentiated (Fig. 7.42).

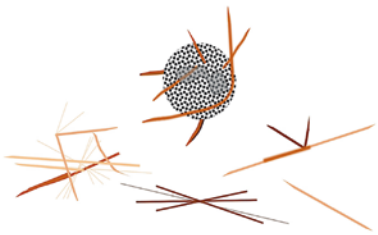


Fig. 7.42 Tyrosine

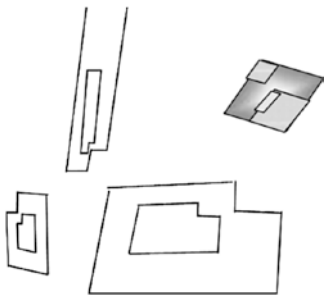


Fig. 7.43 Cholesterol

Note Tyrosine can be confused with nonspecific crystalline needles!

– **Disorder**: severe liver damage

7.6.4 Cholesterol—NR: None

Morphology: Small or large, colorless, angular slabs with angular recesses, mostly superimposed, pH-independent, cholesterol crystals are non-luminescent and have comparatively typical angular contours (Fig. 7.43).

– **Disorder**: renal disease

In the following, amorphous salts and crystals are described whose occurrence depends on diet (e.g. calcium oxalates in tomatoes and rhubarb) on the one hand, but can also be pathological (e.g. hyperoxaluria, hypercalciuria, hyperuricemia).

7.6.5 Urates or Amorphous Uric Acid Salts

Morphology: Brown amorphous salts, similar to grains of sand, can lie completely isolated or densely concentrated in clusters and simulate casts, acidic urine pH. If urates are present in large quantities, urine sediment is reddish-brown in color (brick-dust sediment) (Fig. 7.44).

Urates differ from amorphous phosphates in terms of color and pH value. Color can only be determined in bright-field mode.

Fig. 7.44 Urates

Fig. 7.45 Uric acid crystals

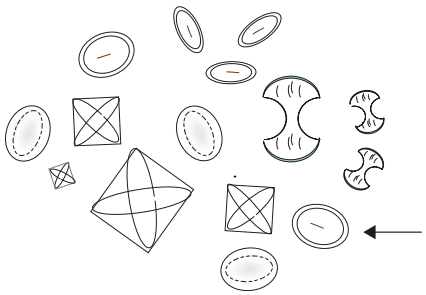


Fig. 7.46 Calcium oxalates. *Arrow*, the round shape of Ca-oxalates should not be confused with biconcave erythrocytes

– **Disorder**: fever, gout

7.6.6 Uric Acid Crystals

Morphology: Yellowish to reddish-brown, varying sizes and highly diverse shapes such as rosettes, rhombic plates, whetstones, dumbbells, barrels, and rods, found in acidic urine (Fig. 7.45).

– **Disorder**: elevated uric acid levels in gout, nephrolithiasis, cytostatic therapy, fever

7.6.7 Calcium Oxalates

– Monohydrate = whewellite = round, oval, hourglass-shaped
– Dihydrate = weddellite = square/envelope-shaped

Morphology: Colorless and translucent in bright-field mode, extremely shiny with a high refractive index, in acidic to slightly alkaline urine, envelope-shaped, round, and oval extremely common, rarely hourglass-shaped (Fig. 7.46).

Fig. 7.47 Amorphous phosphates

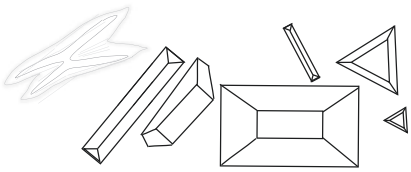


Fig. 7.48 Triple phosphates

Note The round shape of Ca-oxalates should not be confused with biconcave erythrocytes!

– **Disorder**: an increased level of Ca-oxalates can indicate nephrolithiasis, ethylene glycol poisoning (main component of antifreeze).

7.6.8 Amorphous Phosphates (Tricalcium and Trimagnesium Phosphates)

Morphology: White to grey, small crystalline grains of sand, lying individually and in clusters, rarely as plates, in alkaline or mildly acidic urine, not to be confused with urates. Their color can only be determined microscopically in bright-field mode. Macroscopically, sediment is whitish-gray in color (Fig. 7.47).

7.6.9 Triple Phosphates or Ammonium Magnesium Phosphates

Morphology: Colorless, often bright, coffin lid-shaped, rarely thick bars, triangular, or fern leaf-shaped, vary greatly in size, often together with large numbers of bacteria in alkaline as well as old urine (Fig. 7.48). Therefore, it is important to distinguish whether these crystals are only present with increased bacteria (secondary bacterial contamination) or together with increased leukocytes (due to a bacterial urinary tract infection).

Triple phosphates are caused by urea-splitting bacteria (e.g., Proteus).

Disorder: bacterial urinary tract infection, bacteriuria

7.6.10 Calcium Phosphates

Morphology: Comparatively rare, colorless, shiny, rectangular or wedge-shaped crystals that may be arranged individually or in a fan configuration, in rare cases also arranged in plate form, alkaline urine (Fig. 7.49).

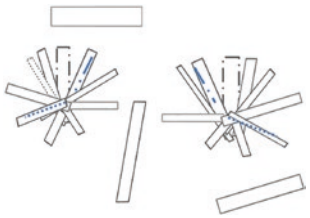


Fig. 7.49 Calcium phosphates

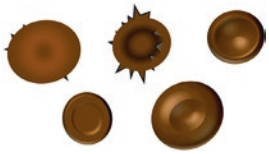


Fig. 7.50 Ammonium urate crystals

– **Disorder**: bacterial urinary tract infection and bacteriuria with urea-splitting bacteria (e.g., *Proteus*).

7.6.11 Ammonium Urate Crystals

Morphology: Yellowish brown spheres of varying sizes, some with small thorns, some with a central indentation (biscuit-shaped), mildly alkaline urine (Fig. 7.50); they occur together with urease-positive bacteria, such as *Proteus*, and in the case of concomitant high uric acid excretion.

– **Disorder**: bacterial urinary tract infection and bacteriuria with urea-splitting bacteria (e.g., *Proteus*).

Note Ammonium urate crystals should not be confused with leucine!

> *Note: The 2,8-dihydroxyadenine or xanthine crystals that resemble ammonium urate crystals (see Hesse 2009) occur only extremely rarely. The microscopic differentiation (bright-field and phase-contrast) of these crystals is not unequivocal. 2,8-Dihydroxyadenine and xanthine urine crystals can be detected using infrared spectroscopy.*

7.6.12 Drug Crystals

The presence of rare and atypical crystals can also be caused by medications. Determining which drug is involved can be challenging, since drug crystals crystallize in different shapes. It is important to make a record of atypical forms of crystallization in the findings, since increased excretion of

drug crystals can also damage the kidneys. For example, sulfonamides, amoxicillin, and indinavir are typical drugs that can cause crystalluria.

7.7 Other Sediment Constituents

7.7.1 Spermatozoa

Morphology: Bright, transparent-appearing head, oval to round in shape, long, thin, dark tail, lying individually or in clusters (Fig. 7.51).

The still vital spermatozoa meander through the HPF. If one misses the tail, it can be difficult to distinguish sperm heads from yeast cells. The head lights up clearly in phase-contrast mode.

7.7.2 Lipid Particles

Morphology: Lipid droplets are extremely shiny, vary in size, and are found individually or clustered both extracellularly and intracellularly (e.g., oval fat bodies, lipid casts, oval fat body casts).

In addition, cholesterol crystallizes into cholesterol crystals.

– **Disorder**: Renal disease with lipiduria and pronounced proteinuria

Note Round lipid particles can also occur as artifacts in urine as a result of the use of ointments and suppositories.

Fig. 7.51 Spermatozoa

7.8 Artifacts

Artifacts arise from:

– Materials used to produce the native specimen (slides, cover glass, gloves)
– The surrounding environment (dust and pollen in the air)
– Sample collection: fibers from patients' clothing, contamination such as feces (particularly in bedridden patients), fatty ointments, suppositories, patient hair.

Moreover, incorrect preparation of the native specimen can result in artifacts (air bubbles, dirty cover glass and microscope slides).

Note It is important to correctly identify and classify artifacts in order to be able to differentiate them from pathological urinary sediment constituents such as erythrocytes, casts, cholesterol, etc.

Comment: A number of artifacts are discussed below. A comparison of artifacts and urinary sediment constituents can be found in Chap. 11, Sect. 11.12.

7.8.1 Fat Droplets

Note Correct identification of fat droplets is important to avoid confusing these with erythrocytes!

Fat droplets in urine are round, highly refractile structures. They can be distinguished from erythrocytes on the basis of their high refractive index and varying sizes (Fig. 7.52).

If no oval fat bodies or fatty casts are seen in addition to fat droplets in urine sediment, these may also have been caused by fatty ointments or suppositories.

Fat droplets alter considerably in terms of brightness and luminosity when the micrometer knob is used.

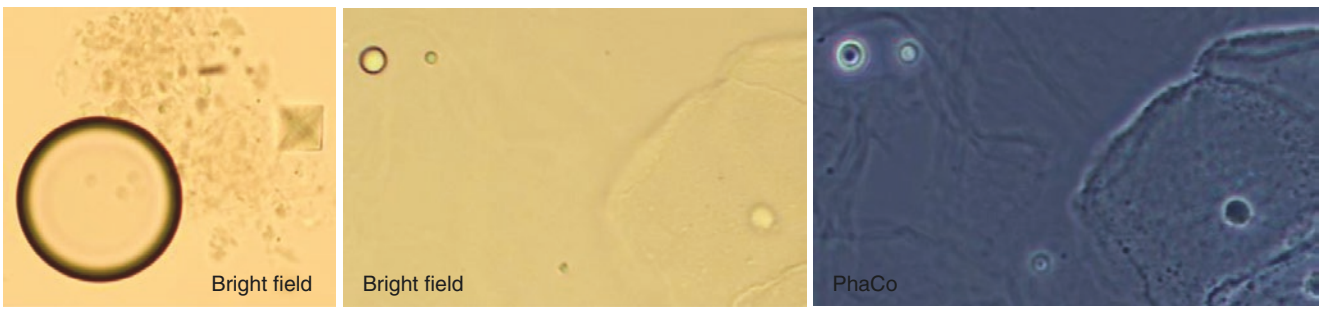


Fig. 7.52 Fat droplets: large and small

7.8.2 Air Bubbles

Note Correct identification of air bubbles is important to avoid confusing these with erythrocytes!

If the native specimen has not been produced correctly, or if the specimen is no longer fresh and has "drawn air", numerous air bubbles of varying sizes may be visible. Air bubbles have a lower refractive index compared with fat droplets (Fig. 7.53).

7.8.3 Glass Fragments

Note Artifacts such as glass fragments must not be confused with cholesterol!

Tiny glass fragments are often found on slides and cover glasses (Fig. 7.54).

7.8.4 Fibers, Dust, and Hair

Note Artifacts such as fibers, dust, hair, and dandruff can simulate casts (Fig. 7.55). All particles can appear either translucent and colorless or colored.

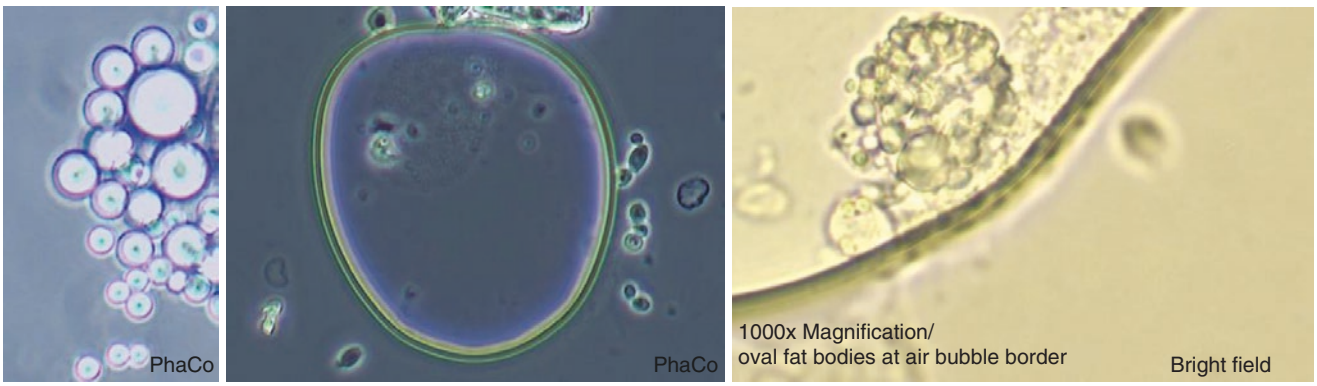


Fig. 7.53 Large and small air bubbles

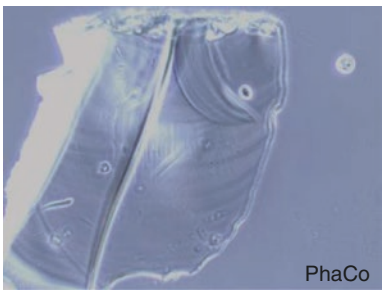


Fig. 7.54 Glass fragment

Fig. 7.55 Fibers, dust, and hair

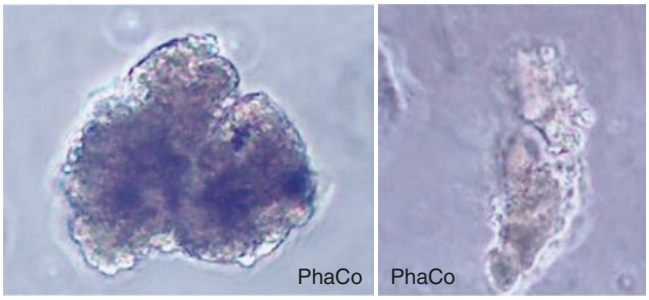


Fig. 7.56 Feces

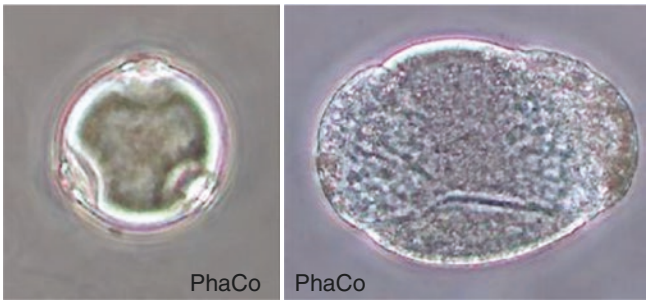


Fig. 7.57 Pollen

7.8.5 Feces

Note Feces should not be confused with urates/uric acid crystals!

Small quantities of feces may be present particularly in the urine of elderly and bedridden patients (Fig. 7.56).

7.8.6 Pollen

Note Artifacts such as pollen can be confused with histiocytes or epithelial cells.

Pollen can be found more frequently in urine sediment specimens in spring due to open windows (Fig. 7.57).

Staining of Urinary Sediment Constituents

8

8.1 Staining Techniques

8.1.1 From the KOVA® System: Staining Solution (Sternheimer-Malbin Solution) (Fig. 8.1)

- **Staining solution**: Ethanol, crystal violet, safranin O, and ammonium oxalate.
- **Shelf-life**: Room temperature, no filtration needed.
- **Performance**: Between 1 and 2 drops of staining solution are added to 1 ml of urine sediment and carefully mixed; microscopy analysis can be performed immediately.
- **Staining result**: See product information leaflet.

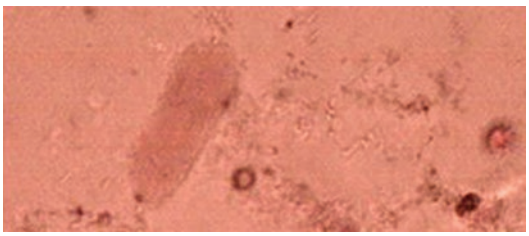


Fig. 8.1 KOVA® stain

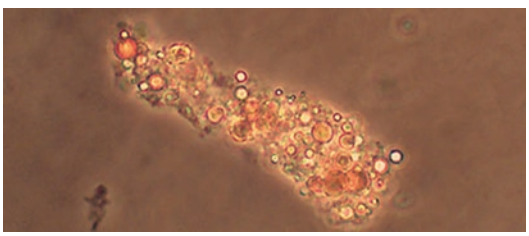


Fig. 8.2 Fat staining with Sudan IV

8.1.2 Fat Staining (Fig. 8.2)

- **Staining solution**: Sudan IV.
- **Shelf-life**: Room temperature.
- **Performance**: Between 1 and 2 drops of filtered staining solution are added to 1 ml of urine sediment and carefully mixed; microscopy analysis can be performed immediately.
- **Staining result**: Fat stains yellowish-red.

8.1.3 Papanicolaou Stain (Complex Stain) (Fig. 8.3)

- For urine cytology and erythrocyte morphology.

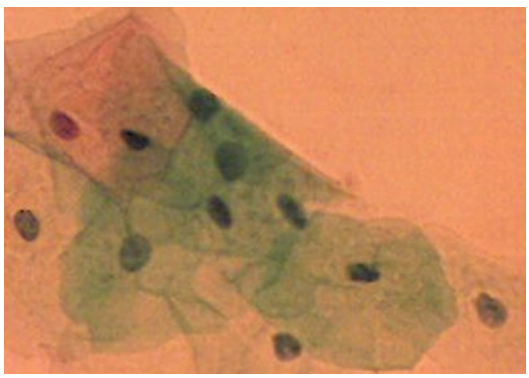


Fig. 8.3 Papanicolaou staining

J. Neuendorf, *Urine Sediment*, https://doi.org/10.1007/978-3-030-15911-5_8

Cell Counting in the Fuchs-Rosenthal Counting Chamber

9

9.1 Discussion: Fuchs-Rosenthal Counting Chamber

Cell counting in the Fuchs-Rosenthal counting chamber is suitable for the precise quantitative determination of erythrocytes and leukocytes in urine. It requires a non-centrifuged urine sample.

Figure 9.1 shows the grid of the Fuchs-Rosenthal counting chamber.

9.1.1 Calculation

Total chamber area	16 mm^2	Chamber area of a group square	1.0 mm^2
Chamber height	0.2 mm	Chamber height	0.2 mm
Total chamber volume	3.2 mm^3 (µl)	Chamber volume of a group square	0.2 mm^3 (µl)
Total number of group squares counted: 5			

$$\frac{\text{Cells counted}}{\text{Areas counted} \quad \text{Chamber height}} = \frac{\text{Cell count}}{5 \times 0.2\mu l} = \text{Cell count / µl urine}$$

$$\left[5 \times 1.0 mm^2\right] \times \left[0.2\mu l\right]$$

Performing the Cell Count

- Add fresh, well mixed native urine to the counting chamber using a dropping pipette.
- Count five group squares.
- The total number of cells corresponds to the number of cells per microliter urine.

9.1.2 Microscope Set-Up

Chamber counting can be performed using both bright-field and phase-contrast methods.

Bright-Field Microscopy By using the 10x objective—with the aperture diaphragm 2/3 closed and (if present) the condenser front lens swung out—the grid plane can be brought into sharp focus. A least square can be set using the 40× objective.

Phase-Contrast Microscopy Same procedure as in bright field; however, the aperture diaphragm on the condenser must be fully opened (otherwise the HPF remains dark). The phase-contrast technique is highly suited to cell counting in the counting chamber.

Cell counting is performed in the five group squares using the 10× or 40× objective. The total number of erythrocytes or leukocytes counted is given per microliter of urine (without further calculation).

9.1.3 Normal Range

- Up to 10 leukocytes/µl
- Up to 3 erythrocytes/µl

9.2 Discussion: Fuchs-Rosenthal Counting Chamber

The Fuchs-Rosenthal counting chamber is particularly suitable for counting fluids with small cell quantities, since it has a chamber height of 0.2 mm and is thus twice as high as other chambers (Fig. 9.2).

J. Neuendorf, *Urine Sediment*, https://doi.org/10.1007/978-3-030-15911-5_9

9.2.1 Sliding on the Cover Glass

Moisten the cover glass with some distilled water and push the thumbs forward over the outer platforms with light pressure (Fig. 9.3). If the rainbow effect of Newton's rings is now visible on the outer platforms, one can be sure that the cover glass is firmly in place.

9.2.2 Filling the Counting Chamber

Using a dropping pipette, a drop of the urine sample (well mixed) is applied between the cover glass and counting chamber. The drop must be precisely distributed over the central platform. In the case of air bubbles or too much urine, the counting chamber needs to be cleaned and re-filled.

9.2.3 Counting Technique

Figure 9.4 shows how the cells marked red in the group square are counted in an L-shape, while the white cells are disregarded.

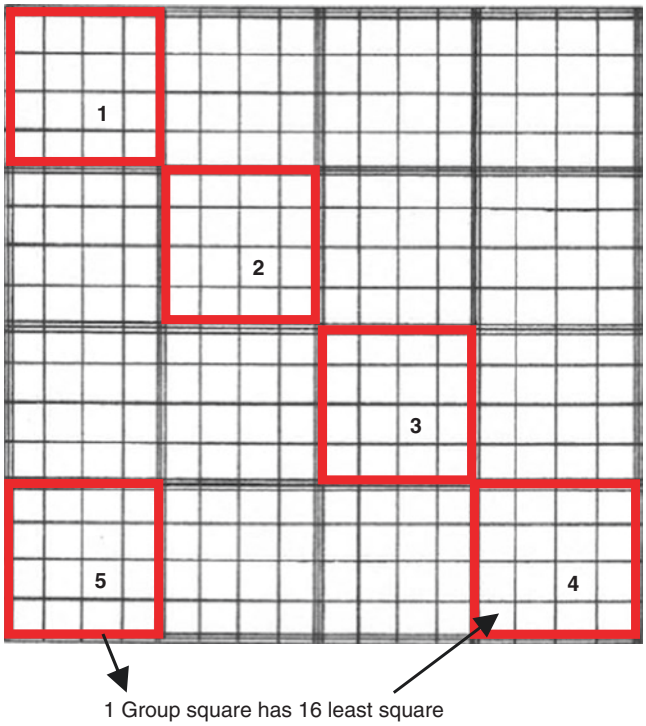


Fig. 9.1 Cell counting in the Fuchs-Rosenthal counting chamber (modified from Rick 1990)

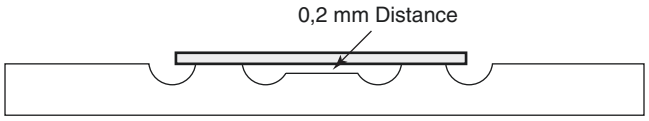


Fig. 9.2 Counting chamber in cross section

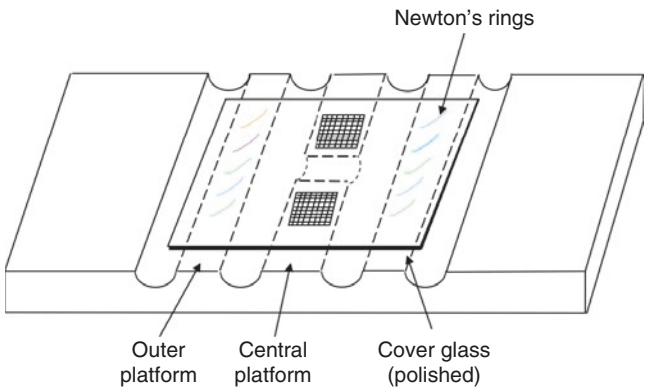
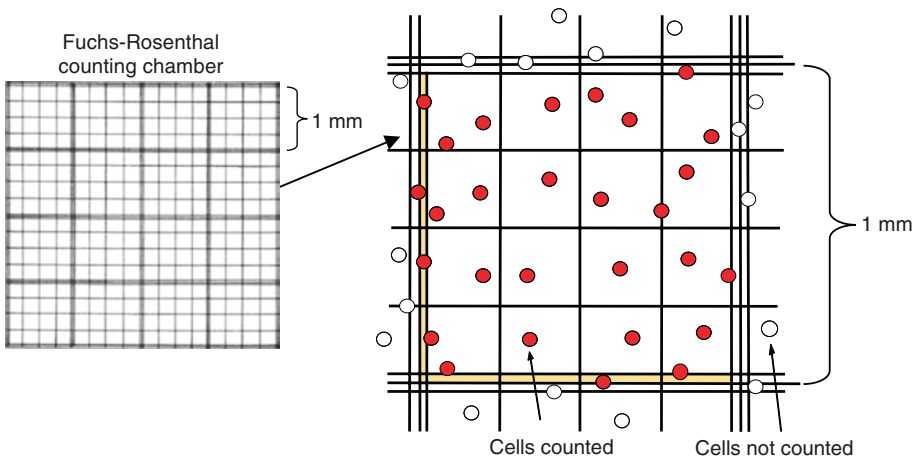


Fig. 9.3 Counting chamber with cover glass and Newton's rings

Fig. 9.4 Counting technique (modified from Rick 1990)



9.2.4 Microscopic Detail of a Group Square/Least Square (Figs. 9.5 and 9.6)

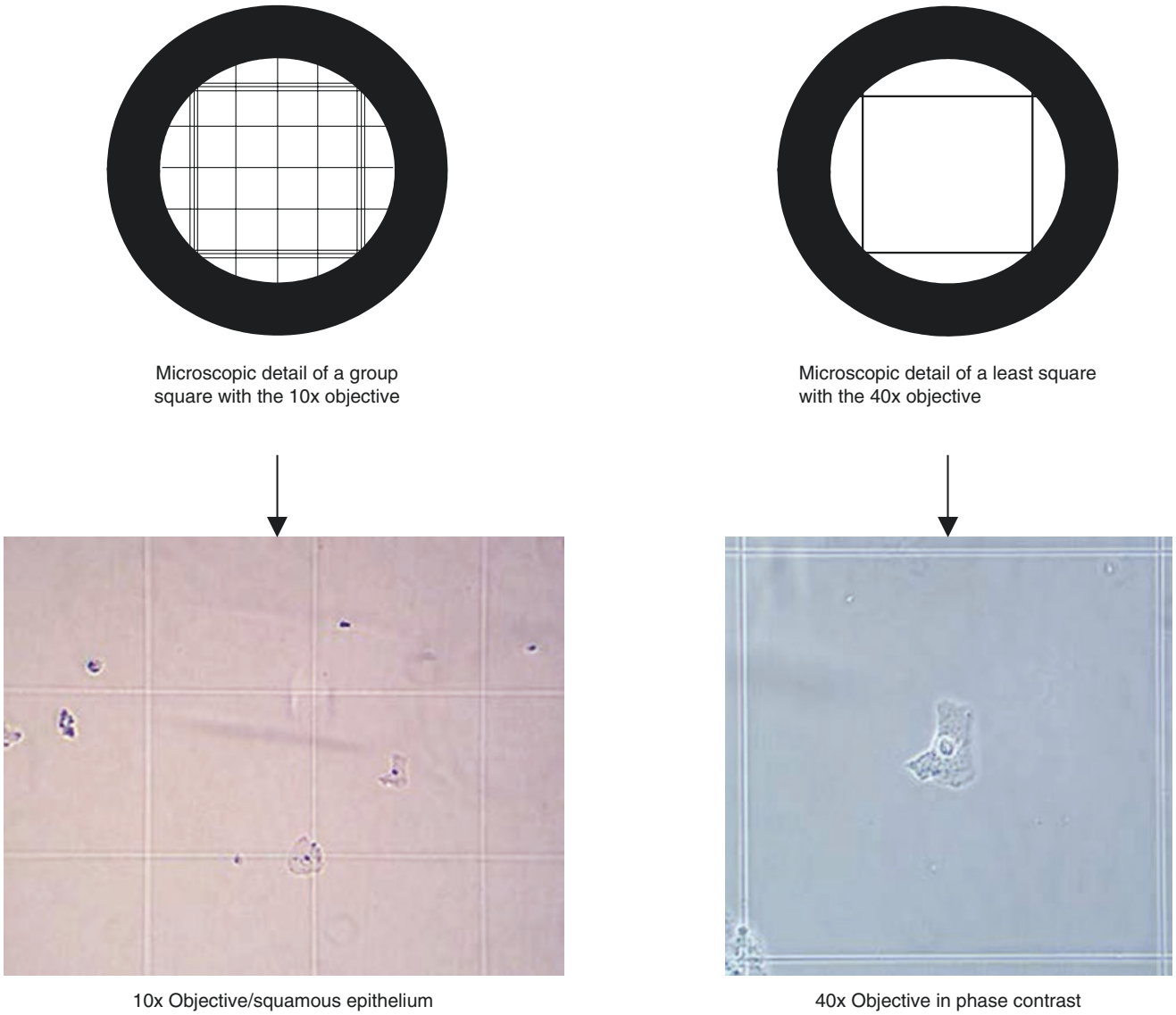


Fig. 9.5 Microscopic detail of a group square/least square

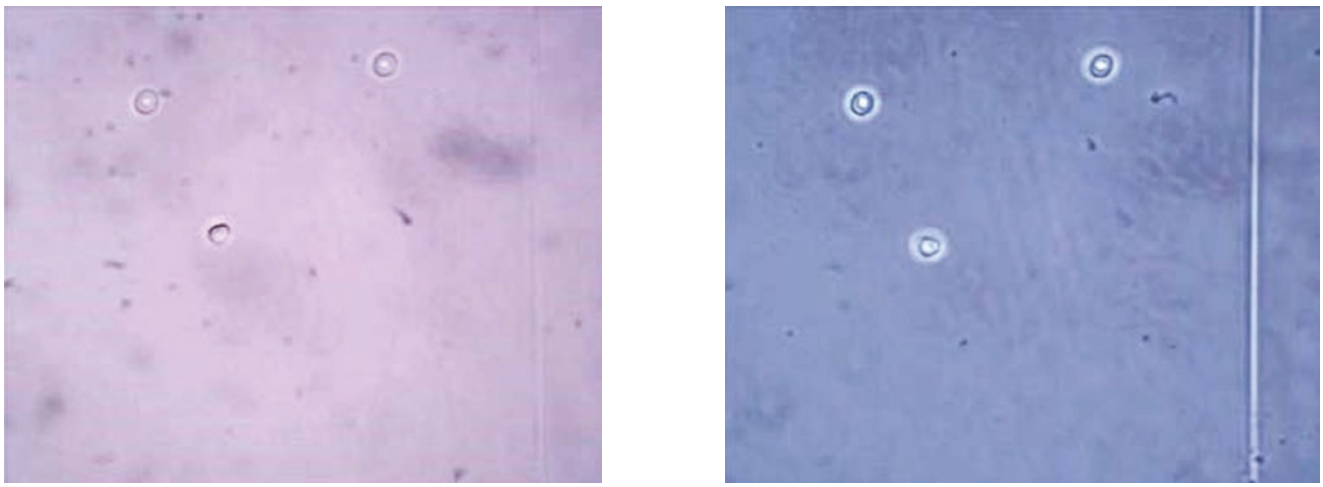


Fig. 9.6 Microscopic details of a least square in bright-field and phase-contrast microscopy

Reference

1. Rick W (1990) Klinische Chemie und Mikroskopie. Springer, Berlin

Hematuria: Laboratory Investigations

10

10.1 Introduction

One of the most important questions in urine diagnosis is whether increased erythrocytes are excreted in urine. Numerous disorders can cause hematuria. It is of enormous diagnostic importance to confirm hematuria indicated on a urine test strip by means of microscopic analysis of urine sediment—and at the same time to differentiate between renal and post-renal hematuria on the basis of erythrocyte morphology.

Unfortunately, difficulties are frequently encountered during microscopic analysis: there are often discrepancies between the evaluation of the urine test strip and the microscopic analysis of urine sediment. These problems are presented and explained in an extensive tabular overview and guidance for troubleshooting is provided.

Hematuria investigation: urine test strip versus urine sediment

Discrepancy: blood testing area on urine test strip: positive and microscopic urine sediment image: Ec: negative

Causes			Troubleshooting tips
Preanalytics: – Fresh/older urine sample		Since erythrocytes lyse in older and alkaline urine samples, they can no longer be differentiated microscopically. The hemoglobin released from the lysed erythrocytes causes a positive result on the blood test area of the urine test strip	Analyze the urine sample within 2 h; avoid exposure to extreme temperatures; avoid long transport distances; use a fresh urine sample to assess hematuria; morphological degeneration of other cells such as epithelial cells and leukocytes indicates an obsolete urine sample

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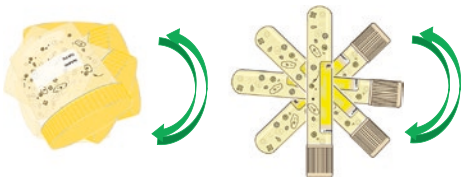
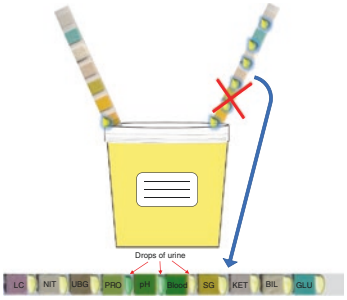
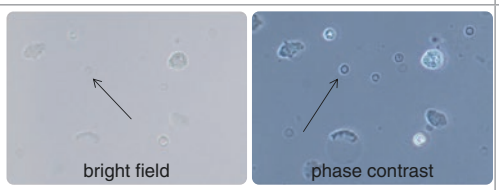
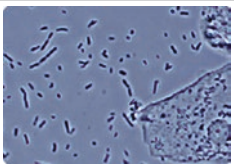
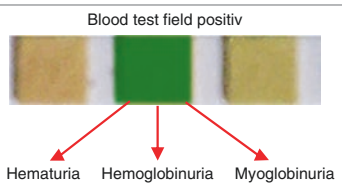
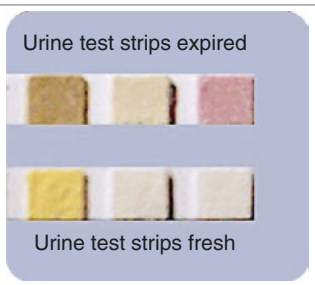
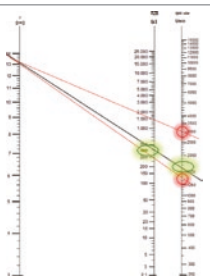
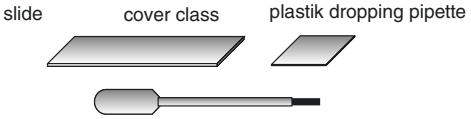
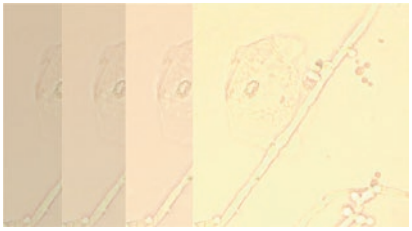
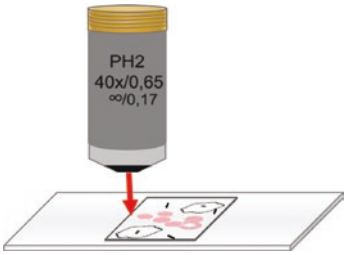
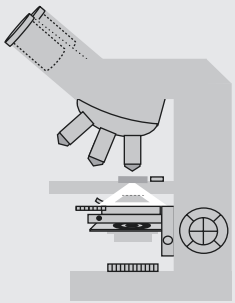
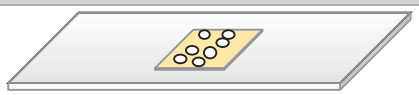
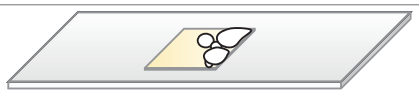
– Mixing		The longer a urine sample stands, the greater the number of erythrocytes and other urine sediment constituents that settle at the bottom of the tube. If the urine sample is not carefully mixed "head-over-head" prior to urine test strip and urine sediment determination, the results will be incorrect	The urine sample test tube must not be slightly agitated, but rather carefully mixed 2–3 times head-over-head. Particularly in the case of suspected microhematuria, a sample that is well mixed head-over-head is the best prerequisite for the correct analysis of the urine test strip and the urine sediment investigation
Urine test strip diagnosis: – Handling		Improper handling of the urine test strip will cause incorrect test strip results. Following immersion of the urine test strip in the urine sample, there is excessive residual urine between the test areas (e.g., a bridge of fluid between pH test area and blood test area: green discoloration of the alkaline pH test area also discolors the adjacent blood test area). This yields a false-positive result for the blood test area	It is essential to ensure correct handling of the urine test strip. Following immersion of the urine test strip in the urine sample, the edge of the test strip must be wiped on the edge of the sample test tube in order to remove excess urine. Remove any excess urine by quickly dabbing the edge of the test strip on a paper towel. Read and follow the package information provided by the urine test strip manufacturer. Perform urine test strip control sample
– Urine pH value > 7 and specific gravity <1.010		At a urine pH value >7 and specific gravity <1.010, predominantly erythrocyte ghosts form, which can lyse rapidly. Erythrocyte ghosts can be easily overlooked in the non-contrasted microscopic bright-field image	Process the urine sample promptly. Erythrocyte ghosts can be better identified using the phase-contrast technique compared to bright-field microscopy. If the phase-contrast mode is not available for microscopy analysis, the microscopic image should be contrasted by closing the aperture diaphragm on the condenser to better visualize erythrocyte ghosts in bright-field mode
– Peroxidase-positive bacteria		The increased presence of peroxidase-positive bacteria such as E. coli, Proteus, and Klebsiellae can cause false-positive reactions on the blood test area	Attention should be paid to the quantity of bacteria during microscopic sediment analysis. Gram-negative rod bacteria such as E. coli, Proteus, and Klebsiellae are among frequent urinary tract pathogens
– Hemoglobinuria – Myoglobinuria		The blood test area on the urine test strip tests positive in the case of erythrocyturia (hematuria), as well as hemoglobinuria and myoglobinuria	Hemoglobinuria investigation → Reduced serum haptoglobin. Myoglobinuria investigation → Increased serum creatine kinase
– Expiry date		If the urine test strips are over their expiry date, false-positive reactions may occur in the blood test area, as well as other test strip areas	Check the expiry date on the urine test strip container the colors of the individual test strip areas can change if the expiry date has been exceeded. Use urine test strips that are within their expiry date

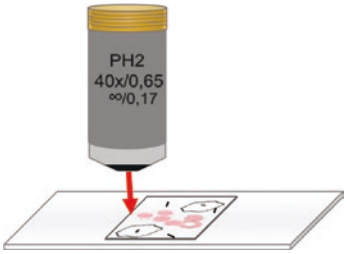
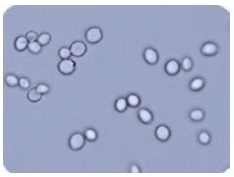
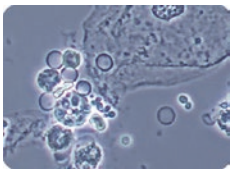
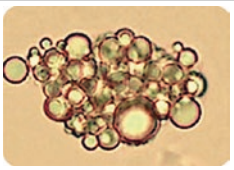
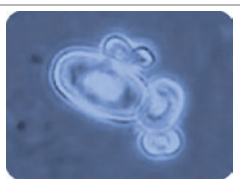
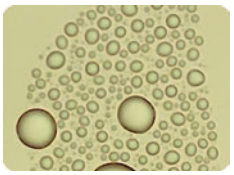
– Storage		The urine test strip container is not closed with the lid after removal of a test strip; humidity and sun exposure reduce urine test strip quality considerably and lead to false results	Open a new urine test strip pack with a valid expiration date, repeat the test, and measure control urine sample. Always remove the appropriate number of test strips for immediate use and securely reseal the container straight away. Do not discard the drying agent in the urine test strip container
– Reaction time		When the individual test strip reaction zones are read visually, the different reading times for the individual test areas are not taken into account. Non-specific color reactions may occur if the reading time of the blood test area is exceeded	Attention must be paid to information on reading times provided on the package leaflet or urine test strip container. Reliable compliance with reading times can be ensured by using urine test strip reading devices
Centrifugation: – Number of revolutions per minute		The urine sample is centrifuged too fast (→ cell lysis) or too slow (→ urine components fail to accumulate optimally in the sediment)	Check the number of revolutions per minute on the centrifuge, e.g. by means of a centrifuge nomogram. Urine samples must be centrifuged at a relative centrifugal force (RCF) of 400 × *g* (European Urinalysis Guidelines 2000). The radius of the centrifuge is needed in order to determine and set the correct number of revolutions (see Sect. 5.4)
Urine sediment preparation: – Decanting		Following centrifugation, the sediment tube is decanted incorrectly. If too much residual urine remains above the sediment, the sediment will be overly diluted upon resuspension. Alternatively, the centrifuged urine sample is decanted twice, following which it no longer contains any sediment. This makes the detection of microhematuria no longer possible	Following centrifugation, hold the urine tube upright for decanting and pour it out with a single motion—count up to 3—then set it upright again. Alternatively, the supernatant can be siphoned off
– Resuspension		Erythrocytes and other urine sediment constituents cannot be correctly determined semi-quantitatively if the sediment has not been carefully mixed and resuspended before preparing the native specimen. Erythrocytes settle in the lower part of the sediment tube	Carefully swing the urine sediment to achieve resuspension. An automatic test tube mixer should not be used to mix the urine sediment. The urine sediment must not be mixed by means of drawing it up several times with a piston pipette or dropping pipette, since this can destroy the sensitive urine constituents
– Native specimen		Dust particles and pollen form artifacts that can significantly impair the microscopic image. Contaminated slides/covers can be the cause of morphological cell changes and cell lysis. Materials for the preparation of the native preparation were stored in an open place	Store slides, cover glasses, and dropping pipettes in a closed place. Pay attention to the expiry date of glass articles (slides and cover glasses)!

Microscope: – Bright-field and phase-contrast technique		Bright-field technique: the microscopic image is too bright with little contrast. Low-contrast erythrocyte ghosts can easily be overlooked	Bright-field microscopy: dim well (use aperture diaphragm lever on the condenser). The phase-contrast technique is able to detect blood shadows as well as low-contrast and colorless urine sediment constituents
– Microscopic level		Microscopic analysis is performed at the wrong microscopy level. Particularly in the case of low cell density (microhematuria), it is difficult to set the correct microscopic level or maintain it	In order to correctly adjust the microscopic level in the native specimen, the edge of the cover glass is brought into focus with the 10× or 40× objective. The phase-contrast technique makes it much easier to adjust the microscopic level. The cleaner the slides and cover glasses are, the easier it is to perform microscopy analysis in the correct level

Discrepancy: Urine test strip, blood testing area: negative and microscopic urine sediment image: Ec: positive



Causes			Troubleshooting tips
Native specimen: – Sample too small quantity		The native specimen was incorrectly prepared. An overly small sample of urine sediment was used. Small air bubbles simulate eumorphic erythrocytes!	Prepare a new native specimen using somewhat more of the urine sediment sample! One needs an approx. 20 µl urine sediment sample for a cover glass measuring 18 × 18 mm
– "Drawn air"		The native specimen is left standing for too long and begins to dry out. In this case, the native specimen has "drawn air". Small air bubbles can simulate eumorphic erythrocytes!	Prepare a new native specimen and perform microscopic analysis immediately
Urine test strip diagnosis: – Vitamin C		Increased excretion of vitamin C (ascorbic acid) in urine affects the blood and glucose test areas and can cause false-negative results	Pay attention to the ascorbic acid test area on the urine test strip

Microscope: – Microscopic level		Incorrect microscopic level set	Using a 10× or 40× objective, bring the edge of the cover glass into focus in order to continue microscopy analysis at the correct level. For example, pollen and small fat particles on the cover glass can simulate eumorphic erythrocytes
Microscopic analysis: – Confusion with yeast cells		Confusion between eumorphic erythrocytes and yeast cells	Morphologically, eumorphic erythrocytes and yeast cells can look very similar. Therefore, pay attention to cell positioning: yeast cells can lie in clusters, form chains, and/or typically assume "mother–daughter asymmetry." The nucleus of a yeast cell can sometimes also be differentiated. The phase-contrast technique facilitates the morphological differentiation between yeast cells and eumorphic erythrocytes
– Confusion with chlamydospores		Confusion between eumorphic erythrocytes (erythrocyte ghosts) and chlamydospores	Erythrocyte ghosts and chlamydospores look very similar and are difficult to differentiate. Chlamydospores are always found together with fungal hyphae
– Confusion with fat droplets		Confusion between eumorphic erythrocytes and fat droplets	Make good use of the micrometer knob! Highly shiny round constituents are typical of fat particles. Fat can enter the urine sample as an artifact (ointments or suppositories) or occur, e.g., in nephrotic syndrome [in the form of oval fat bodies, lipid casts, and extracellular fat droplets (individually or in clusters)]
– Confusion with round/oval calcium-oxalates		Confusion between eumorphic erythrocytes and round/oval calcium-oxalates	Constantly use fine focus adjustment on the micrometer knob: crystals light up considerably, in contrast to erythrocytes. Crystals exhibit significant size difference, in contrast to eumorphic erythrocytes. Even the consumption of, e.g., tomato soup or rhubarb can cause small amounts of calcium-oxalates to be excreted in urine
– Confusion with air bubbles		Confusion between eumorphic erythrocytes and air bubbles	In contrast to eumorphic erythrocytes, air bubbles differ considerably in size

Part II

Urinary Sediment Constituents in Bright-Field and Phase-Contrast Microscopy

11

Electronic supplementary material The online version of this chapter (https://doi.org/10.1007/978-3-030-15911-5_11) contains supplementary material, which is available to authorized users.

J. Neuendorf, *Urine Sediment*, https://doi.org/10.1007/978-3-030-15911-5_11

11.1 Eumorphic Erythrocytes

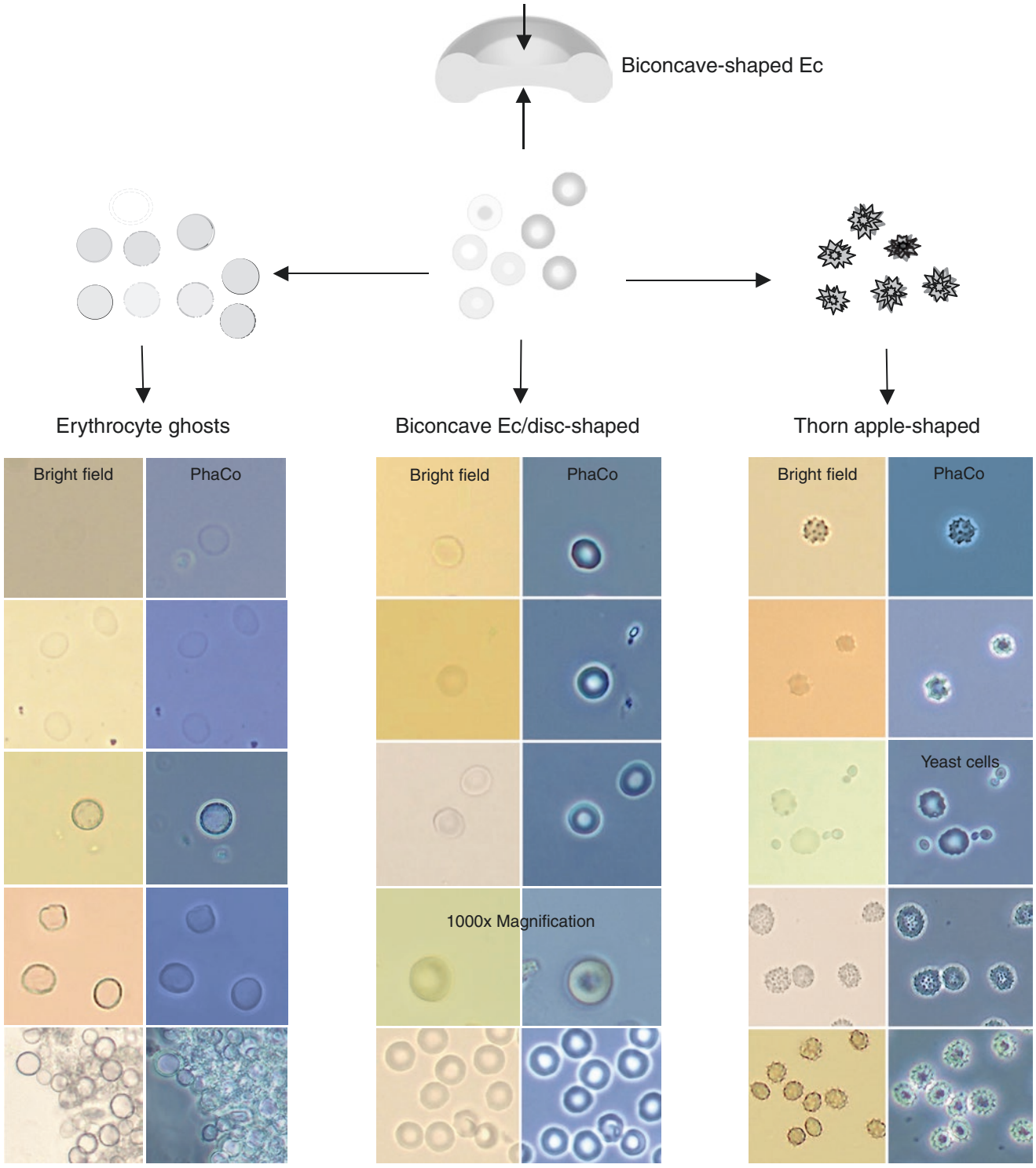
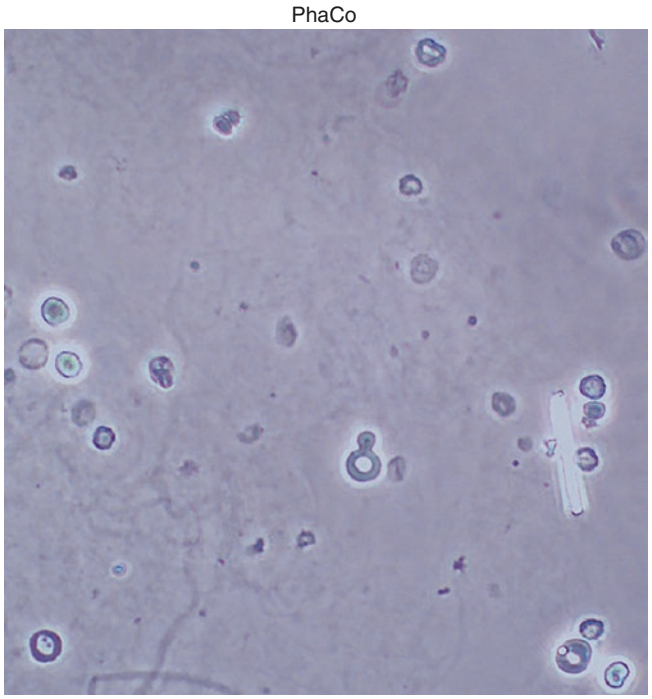


Fig. 11.1 The various morphological details of eumorphic, i.e., normal, erythrocytes. These must not be confused with dysmorphic erythrocytes. Erythrocyte morphology can be particularly well visualized in the phase-contrast image

11.2 Hematuria



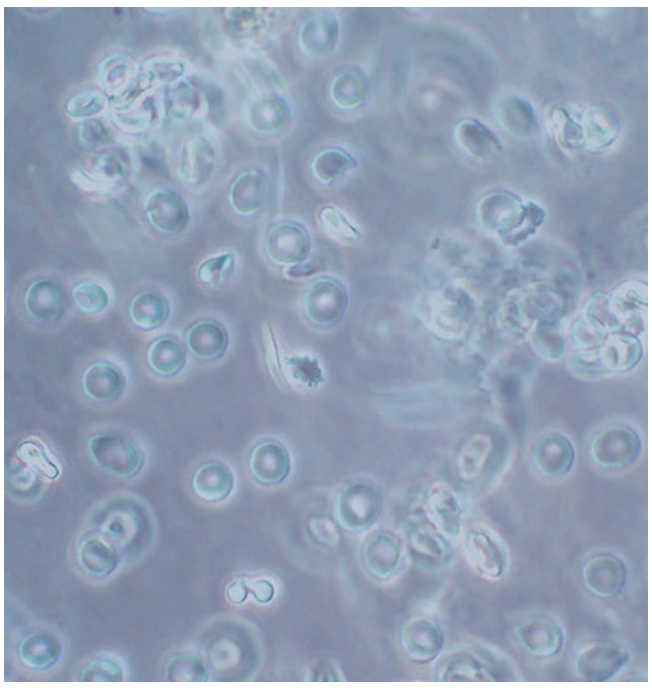


Fig. 11.2 In general, renal hematuria (or erythrocyturia) with predominantly dysmorphic erythrocytes is differentiated from postrenal hematuria with mostly eumorphic erythrocytes. This rule does not apply to renal tumors, which can be accompanied by eumorphic hematuria

11.2.1 Erythrocyte Accumulations

Coagulated blood in urine sediment can sometimes simulate erythrocyte casts—therefore, one should pay attention to red clumps in the urine sediment tube and in the native specimen at the macroscopic stage.

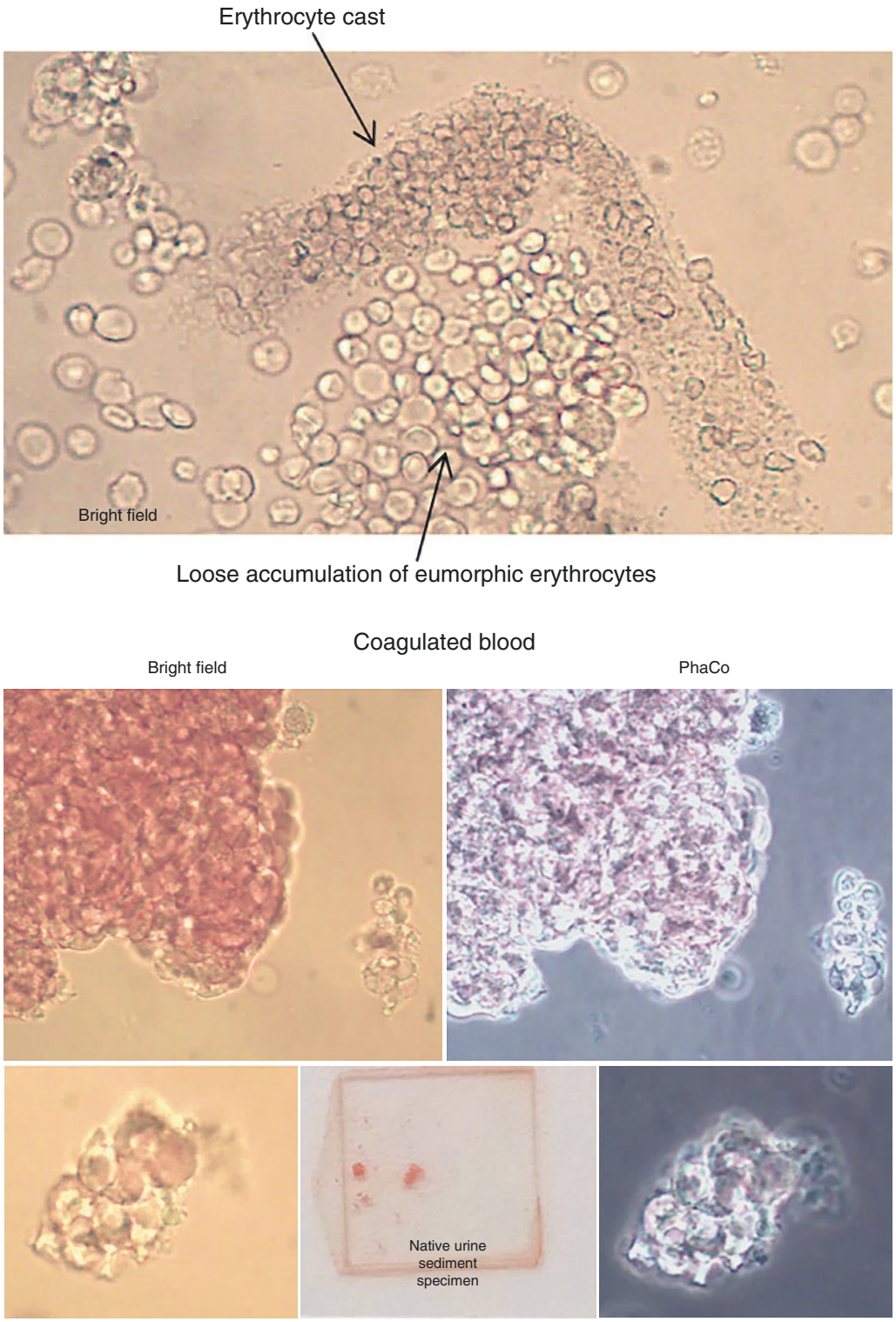


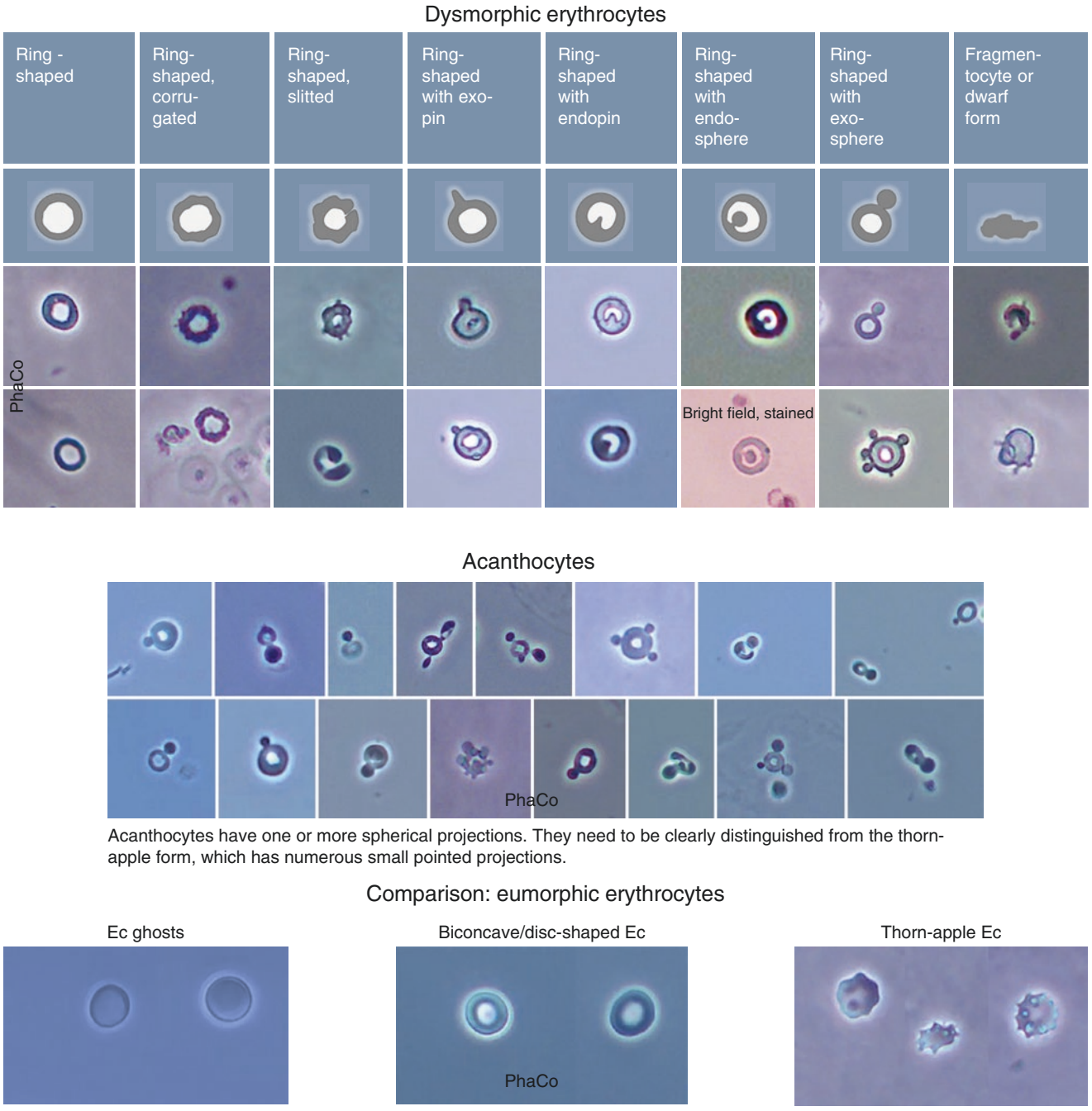
Fig. 11.3 Erythrocyte cast and loose accumulation of eumorphic erythrocytes (*top*), coagulated blood (*bottom*)

11.3 Dysmorphic Erythrocytes and Acanthocytes

This tabular overview shows typical dysmorphic characteristics (Classification see Thiel 1986). Dysmorphic erythrocytes are predominantly microcytic and therefore usually smaller than eumorphic erythrocytes.

The differentiation of dysmorphic erythrocytes is straightforward using the phase-contrast technique.

The acanthocyte—a ring-shaped erythrocyte with at least one typically external exosphere or, rarely, an internal endosphere—represents a special form of dysmorphic erythrocyte.



Acanthocytes have one or more spherical projections. They need to be clearly distinguished from the thorn-apple form, which has numerous small pointed projections.

Fig. 11.4 Dysmorphic erythrocytes and acanthocytes compared to eumorphic erythrocytes

11.4 Yeast Cells and Fungal Hyphae

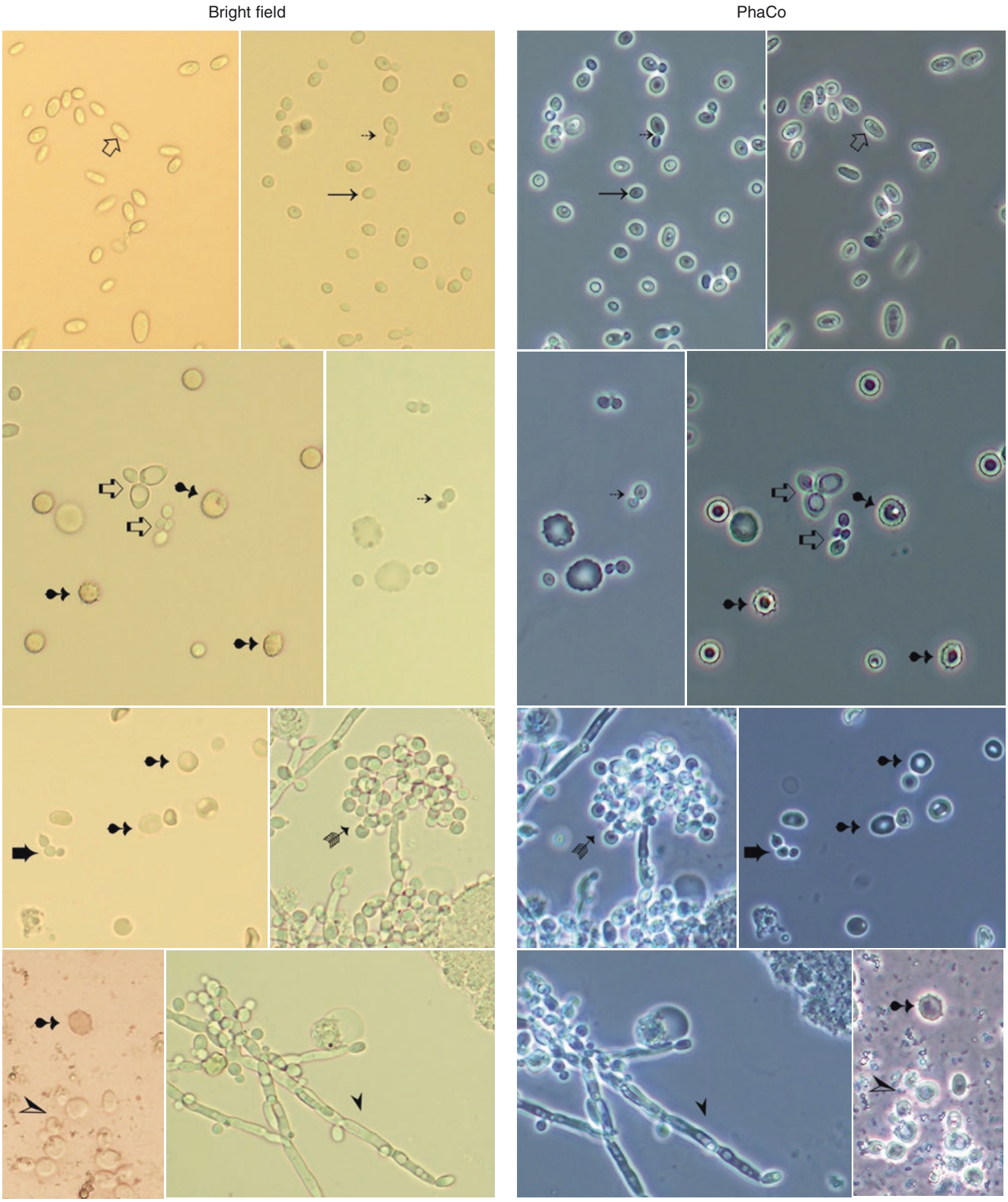


Fig. 11.5 Yeast cells can be round (→) or oval (⇨) in shape. They can be found individually, in pairs in "mother–daughter asymmetry" (one smaller and one larger yeast cell ⇢), in chains (➡) or clusters (➽), and sometimes together with fungal hyphae (➤). Yeast cell size varies greatly (marked anisocytosis ➪). Large yeast cells (➢) are easily confused with erythrocytes (➧)

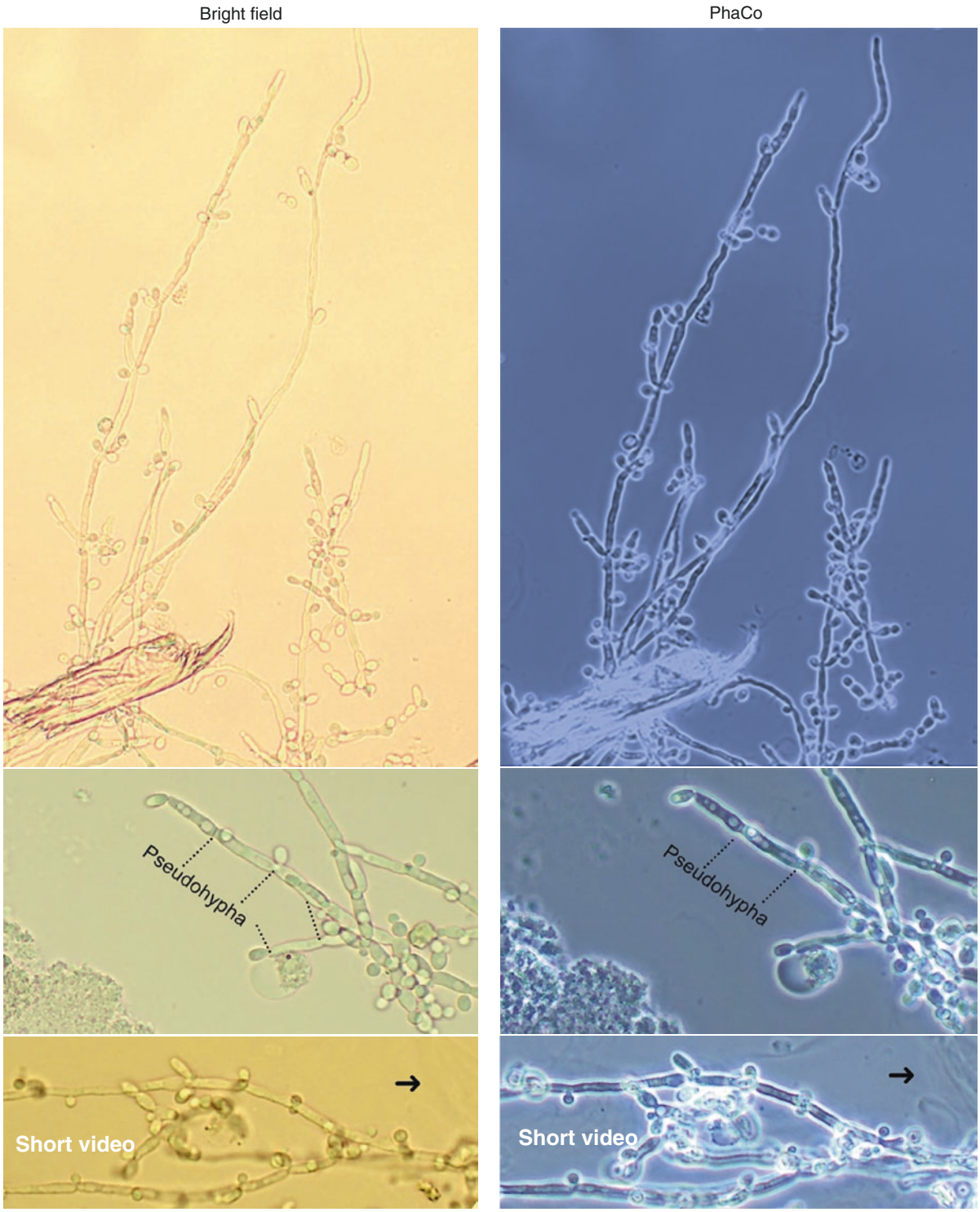


Fig. 11.6 Fungal hyphae have a double-walled tubular shape and can therefore be distinguished from hyphal structures such as bacterial hyphae and mucus threads (➜). (see Videos 11.1 and 11.2)

11.4.1 Yeast Cells, Fungal Hyphae, and Erythrocytes: 1000× Magnification

The various morphological details of eumorphic erythrocytes (➙ thorn-apple form, ⇨ biconcave form), as well as yeast cells (⇛) and fungal hyphae (☛), are clearly visible here.

Yeast cells with visible nucleus and erythrocytes (➻): the nuclear segments (➢) in the yeast cells are clearly identifiable as dark spots.

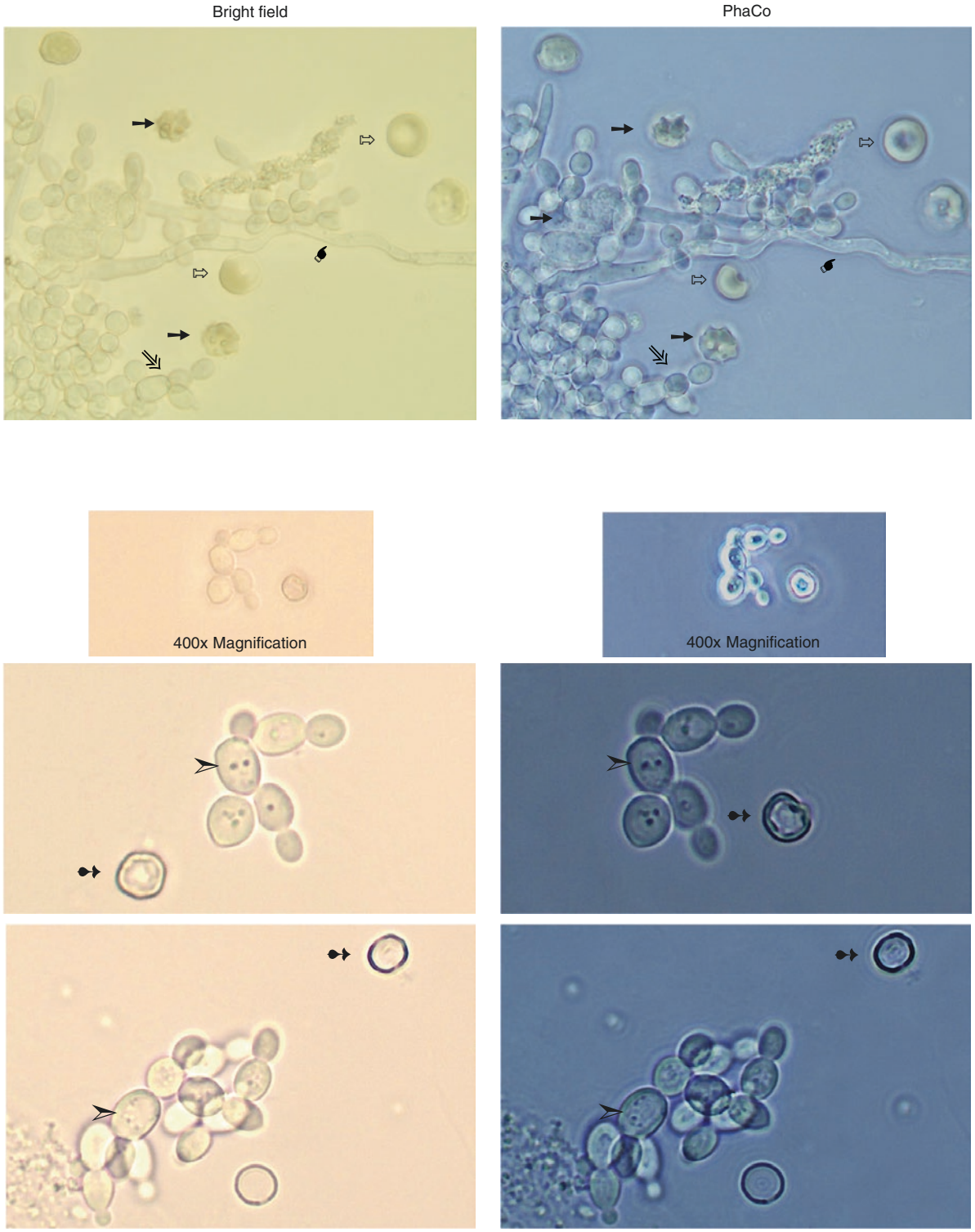


Fig. 11.7 Yeast cells, fungal hyphae and erythrocytes at 1000× magnification

11.4.2 Cluster Formation: Yeast Cells and Fungal Hyphae

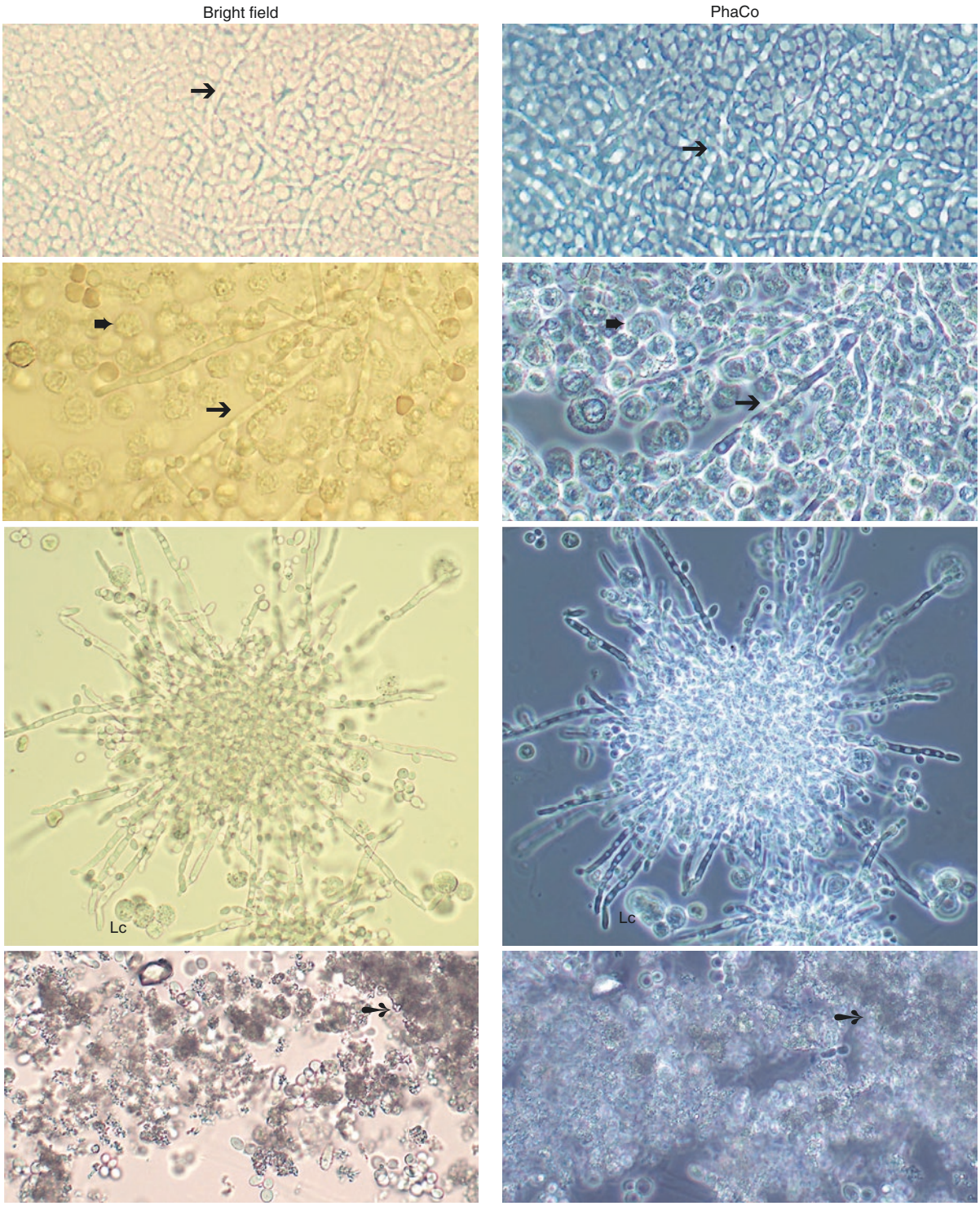


Fig. 11.8 The densely clustered round yeast cells that are permeated by fungal hyphae (➔) are characteristic. Macroscopically, small white nuggets are visible in the urine sediment. Fungal hyphae lie between leukocytes (➨). Since yeast cells and amorphous urates (⇢) condense the PhaCo image, differentiation is only possible in bright-field mode

11.4.3 Yeast Cells with Chlamydospores

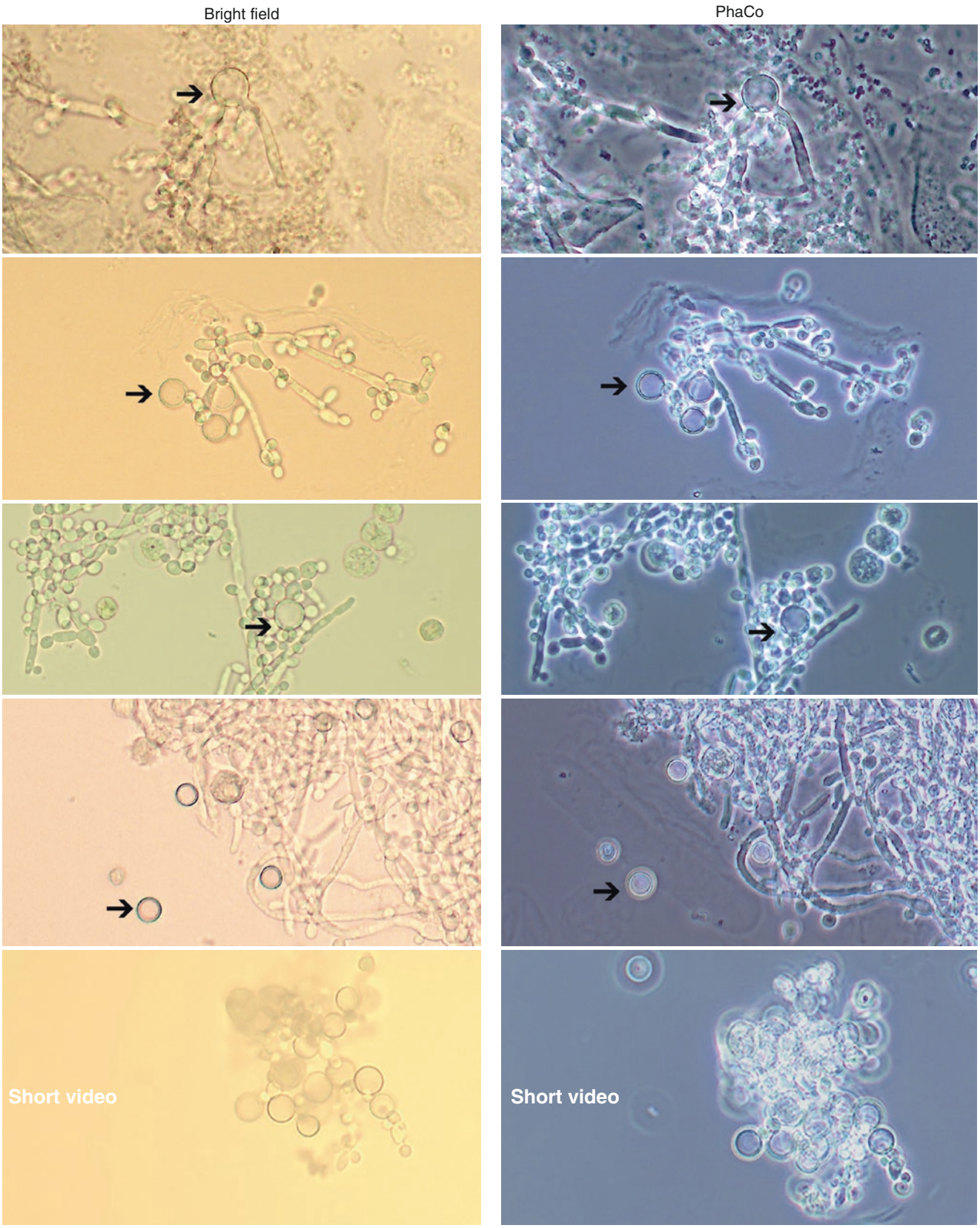


Fig. 11.9 Chlamydospore (➔) seen as a round, double-walled disc that grows terminally on the pseudomycelium. Chlamydospores are easily confused with erythrocytes (erythrocyte ghosts). (see Videos 11.3 and 11.4)

11.4.4 Comparison: Yeast Cells (Mother–Daughter Asymmetry)–Acanthocytes

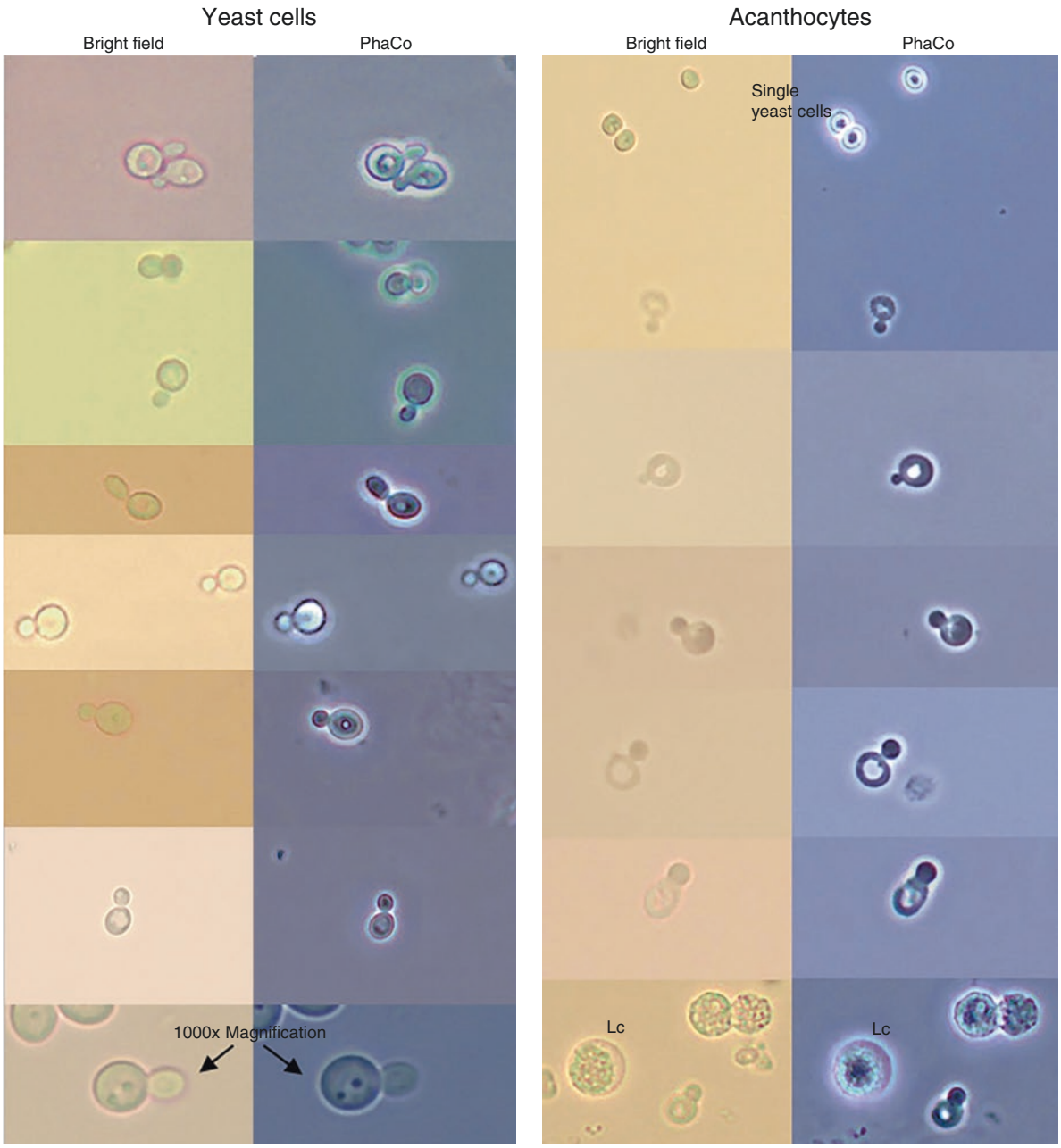


Fig. 11.10 In particular the mother–daughter asymmetry of spherical yeast forms is easily confused with acanthocytes

11.4.5 Bacteria, Fungal Hyphae, and Mucus Threads

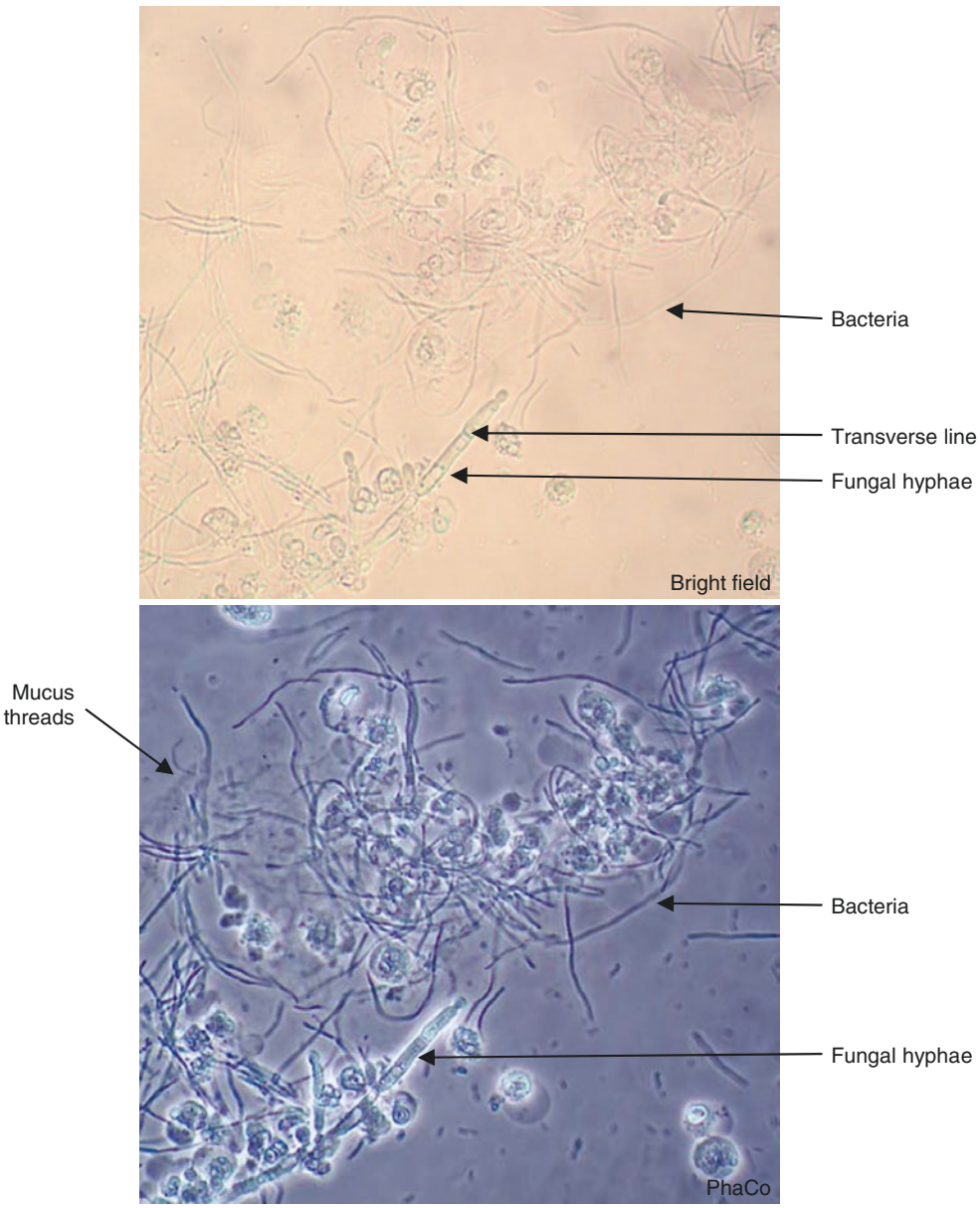


Fig. 11.11 Various elongated shapes: fungal hyphae form tubes that are broken up by transverse lines; mucus threads, which are highly bizarre and irregular, are only visible in PhaCo; long bacteria appear as dark threads

11.5 Leukocytes (Granulocytes)

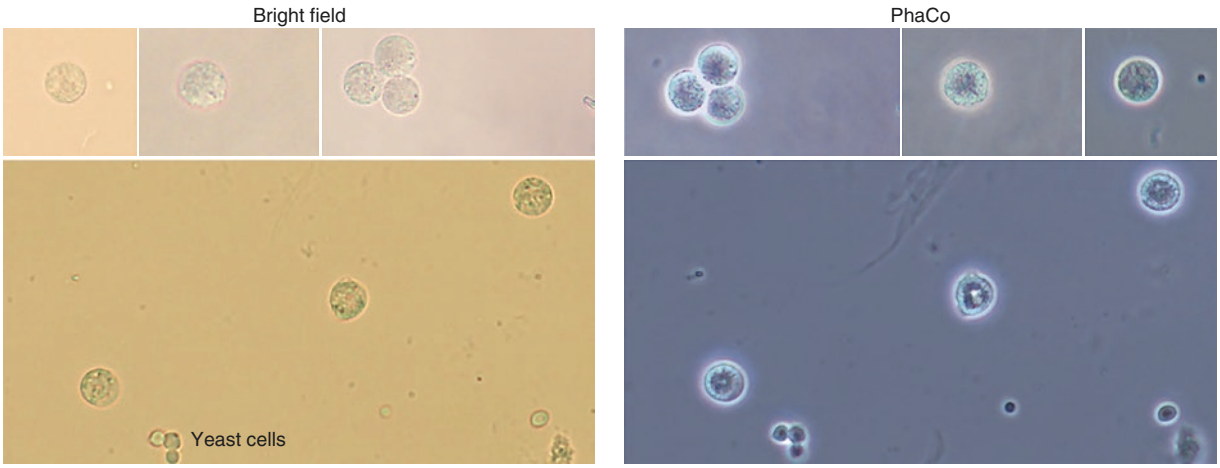


Fig. 11.12 Small-cell leukocytes (granulocytes). Small-cell leukocytes have a characteristic granular surface, are always larger than erythrocytes, and somewhat smaller than renal epithelial cells

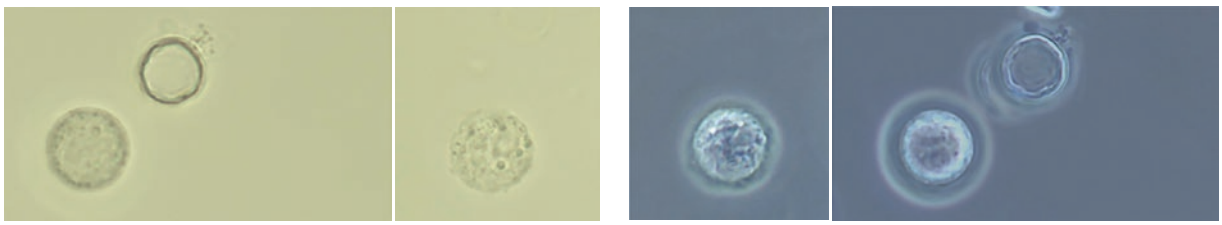


Fig. 11.13 Comparison of leukocytes with eumorphic erythrocytes at 1000× magnification

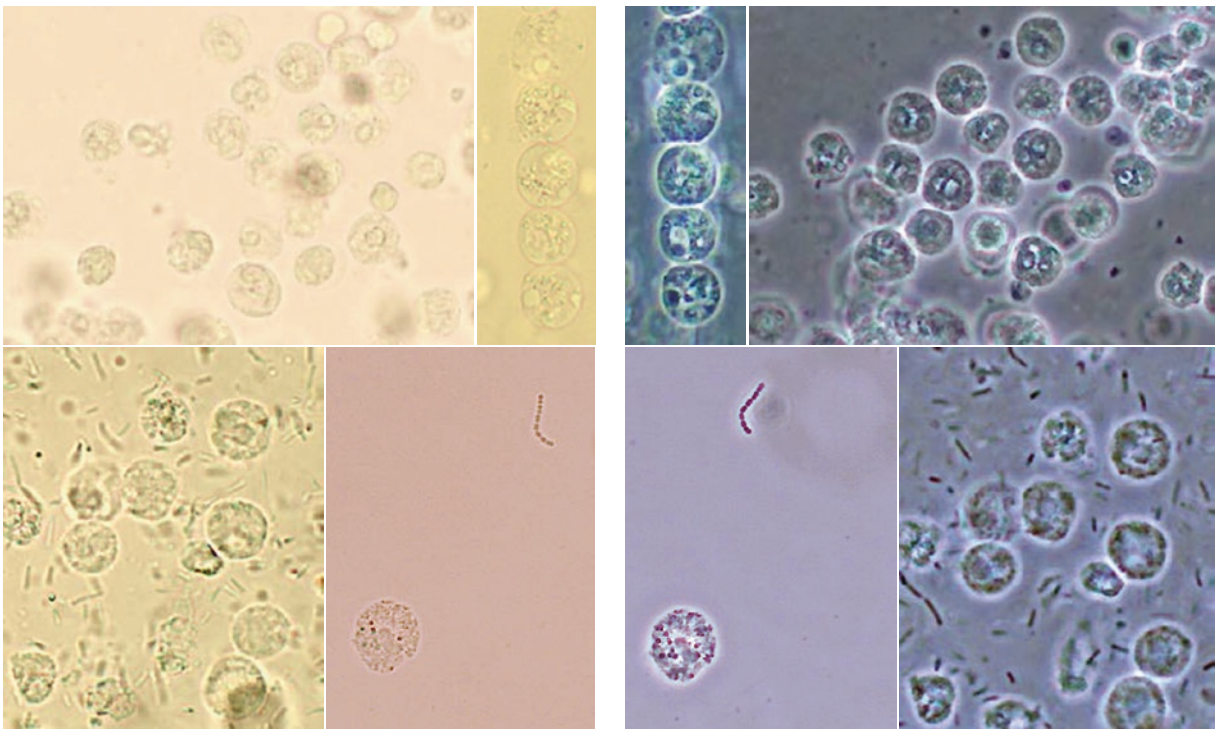


Fig. 11.14 Large leukocytes (granulocytes) with visible nucleus

11.5.1 Old Leukocytes

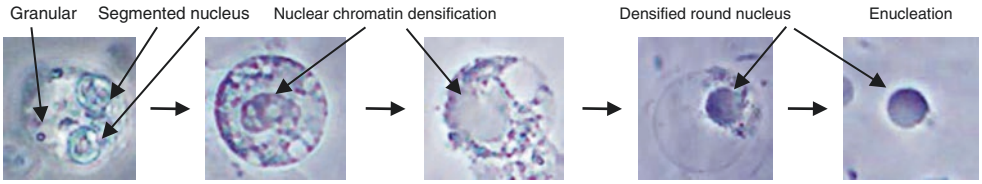


Fig. 11.15 Leukocyte (segmented granulocyte) cell-aging process

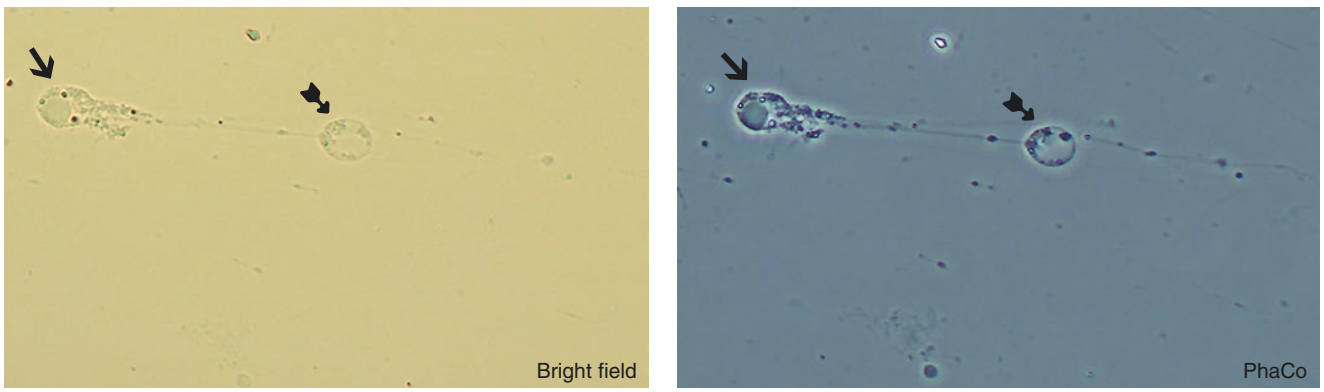


Fig. 11.16 One can see that the leukocyte nucleus (➔) has separated from the cytoplasm (➸)

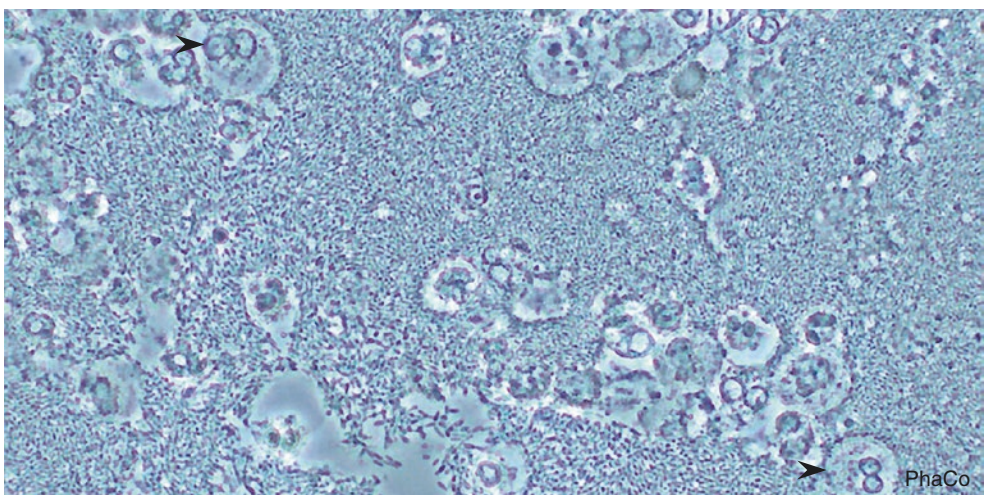


Fig. 11.17 Leukocyte morphology is subject to significant changes if the urine sample is older than 2 h and/or has an alkaline pH value. Enlarged leukocytes (➤) with clearly visible segmented nuclei can be identified on the image between the massive number of bacteria

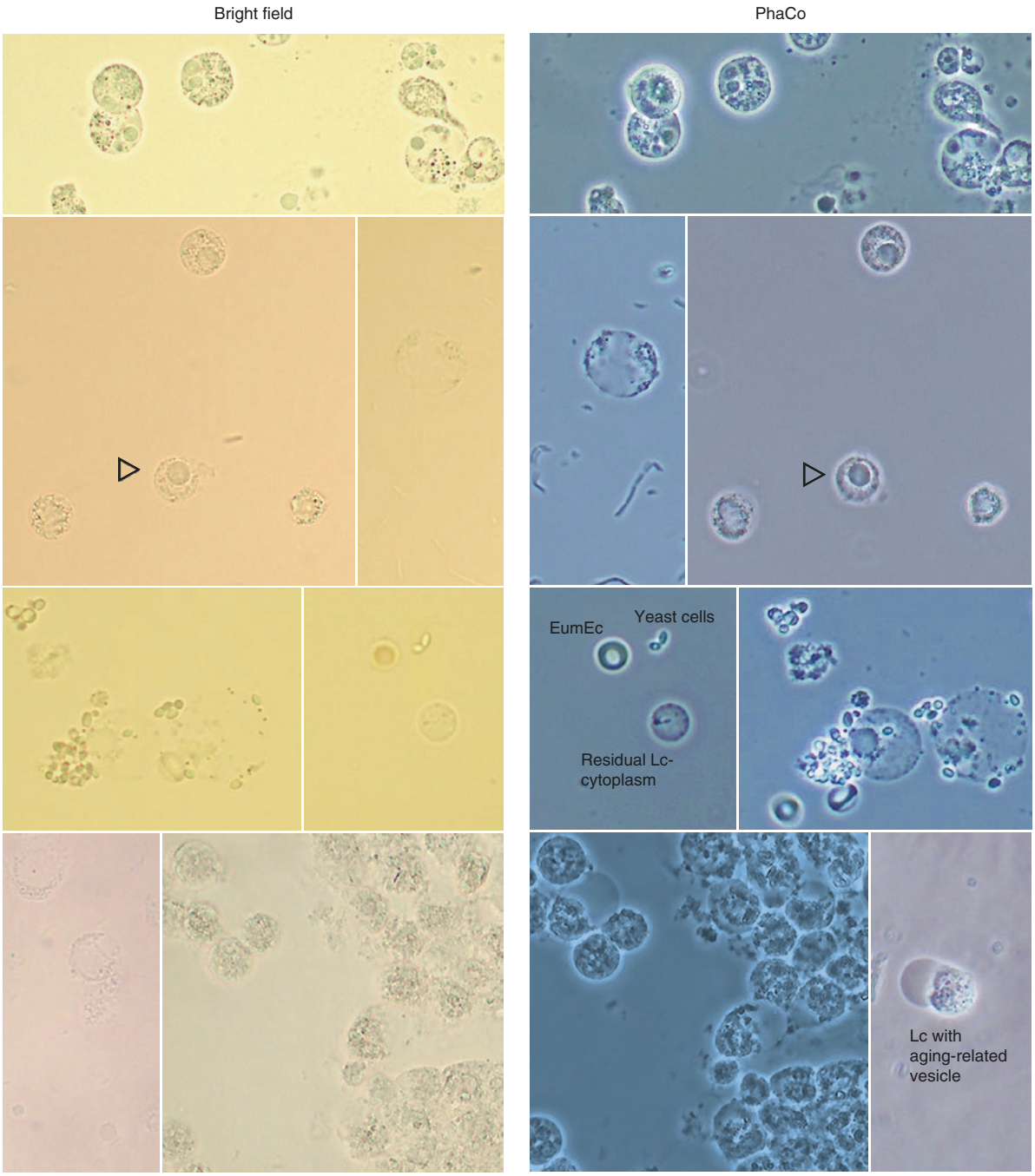


Fig. 11.18 Old leukocytes alter their appearance considerably and lyse. If the urine sample is old and has an alkaline pH value, there may be large discrepancies between the leukocyte test result on the urine test strip and microscopic analysis. Leukocytes with pycnotic, round nuclei (▷) can be easily confused with small-cell epithelial cells such as renal epithelial cells and decoy cells

11.5.2 Elongated Leukocytes

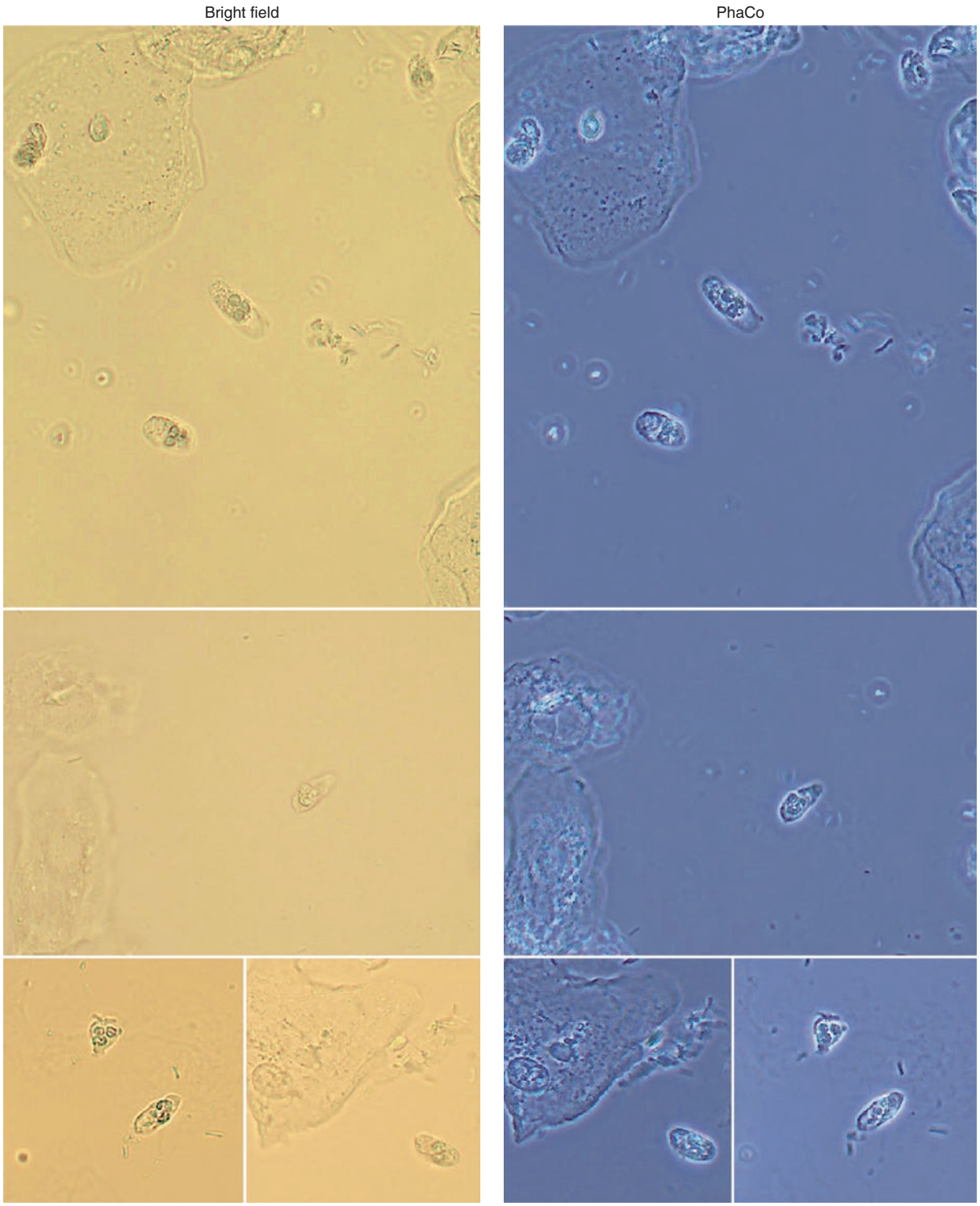


Fig. 11.19 Rare: the typically segmented leukocyte nuclei in the elongated cell form are particularly well visualized in bright-field mode—and must not be confused with deep urothelial cells. The condenser should be slightly closed for bright-field image contrast

11.5.3 Leukocyte Accumulations: Pyuria, Casts, and Clusters

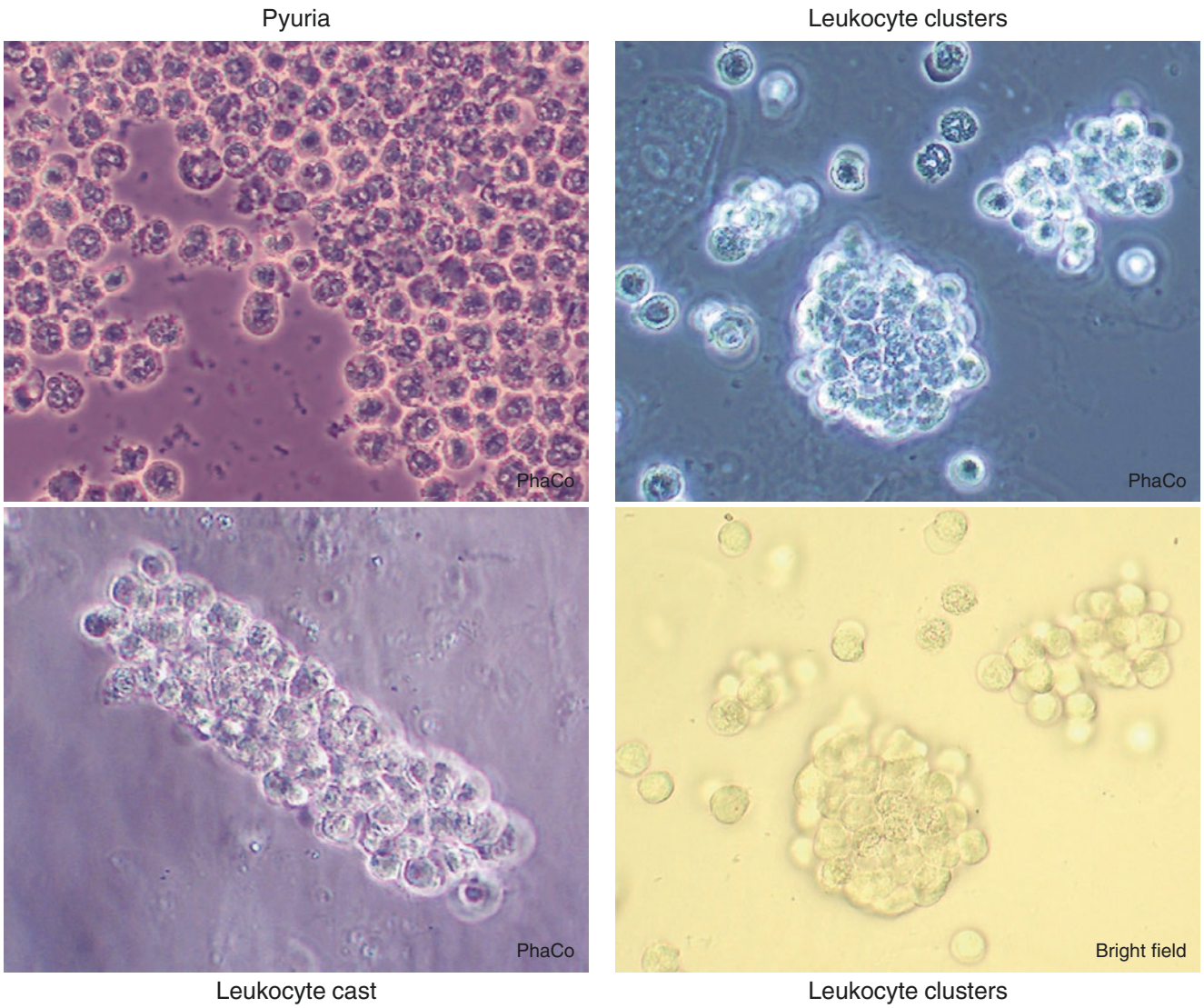


Fig. 11.20 Leukocyte casts are of renal origin and need to be distinguished from irregular leukocyte accumulations. In the case of pyuria, urine is whitish and the sediment highly viscous

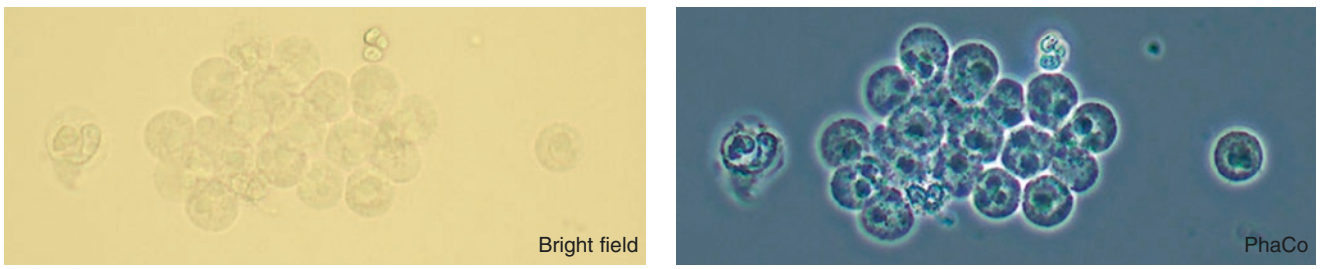


Fig. 11.21 Leukocytes with more rounded cell nuclei are visible in this cell accumulation

11.5.4 Comparison: Thorn Apple-Shaped Erythrocytes with Small-Cell Leukocytes

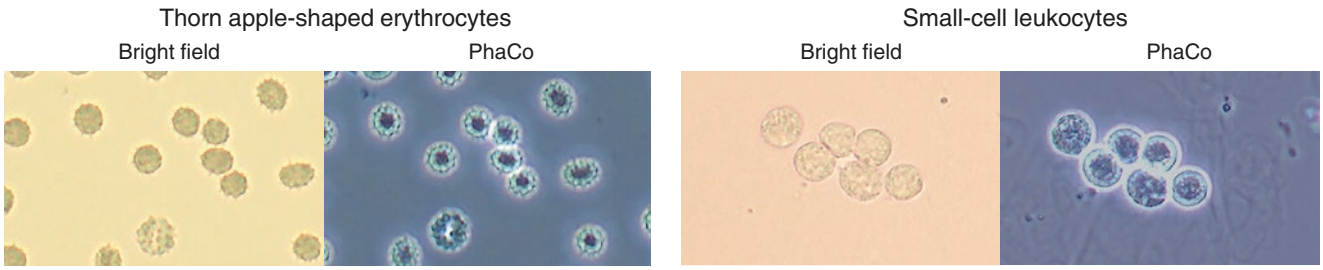


Fig. 11.22 Since the surface of the thorn apple-shaped erythrocyte appears dark and granular, its differentiation from small-cell leukocytes may be challenging. In addition to evaluating the different cell sizes, the different contours of the two cell types also helps in correct differentiation: due to the pointed projections of the thorn apple-shaped erythrocytes, an uneven contour can be visualized, while the leukocyte contour appears smooth throughout

11.5.5 Comparison: Fresh Native Specimen and Old Native Specimen from the Same Urine Sample

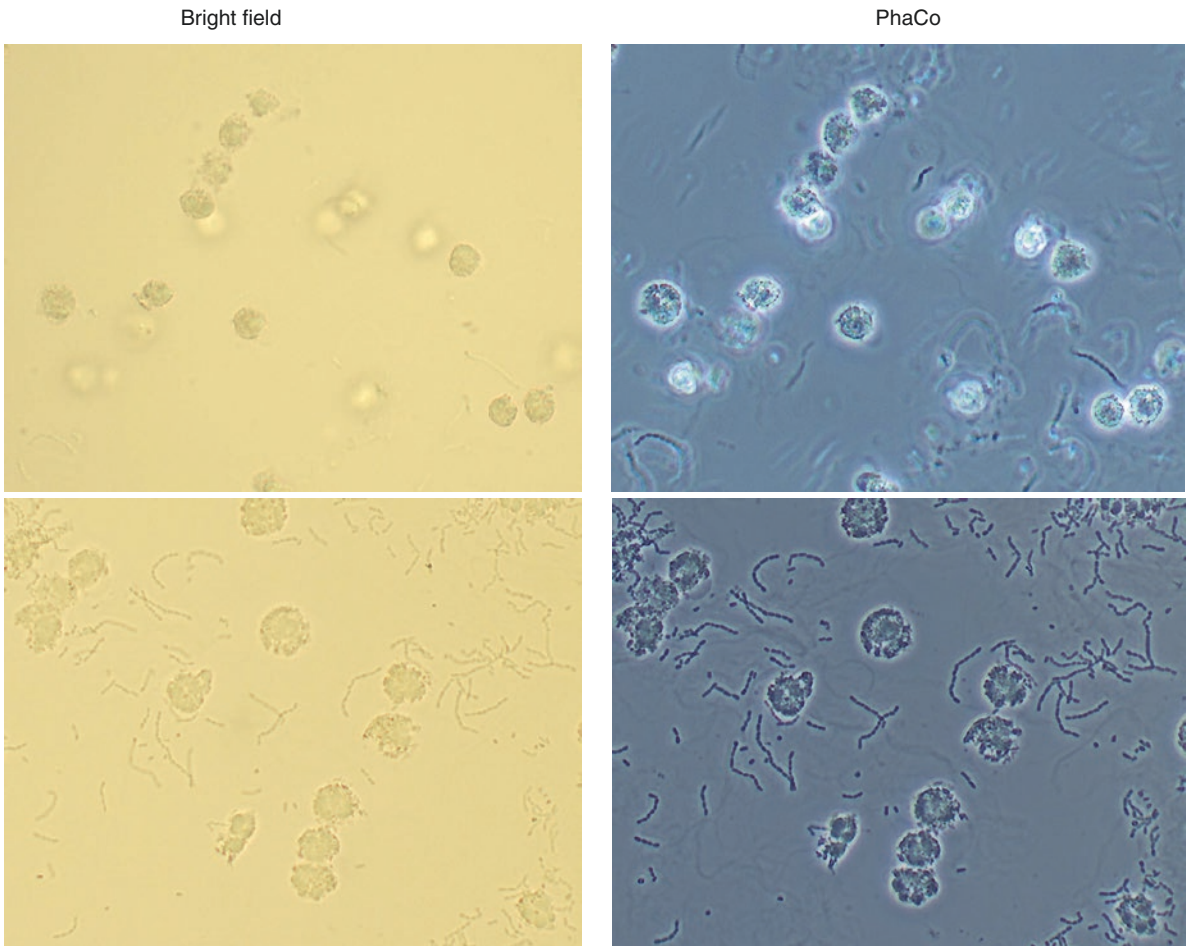


Fig. 11.23 Fresh native specimen: small-cell leukocytes are predominantly present (top). Older native specimen: leukocytes are irregularly enlarged and in some cases the nucleus is visualized (bottom)

11.5.6 Leukocytes with Phagocytized Yeast Cells

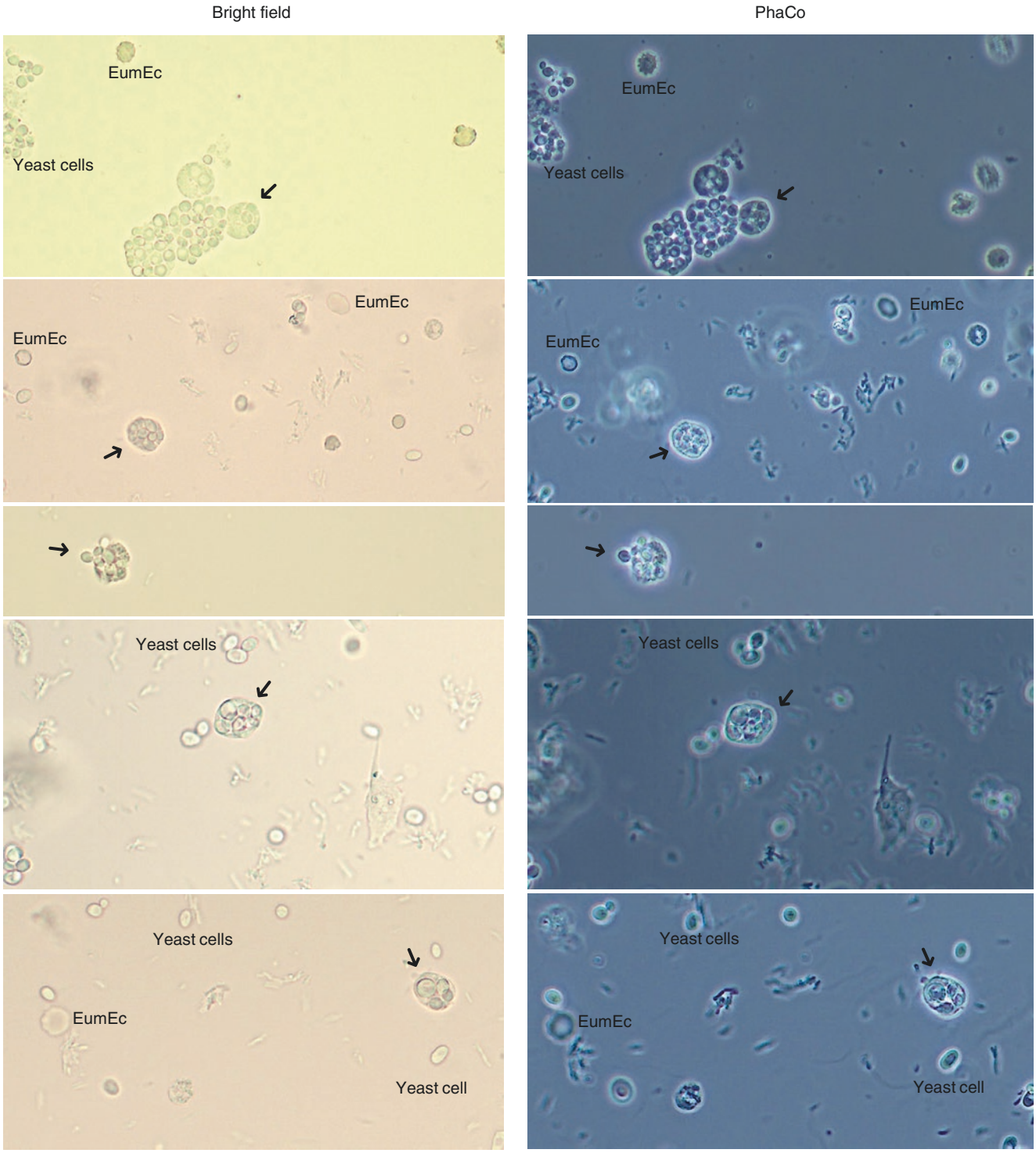


Fig. 11.24 Cellular storage of yeast cells (➜) in the leukocytes is readily visualized. Due to intracellular structure density, yeast cells are particularly well visualized on bright-field images

11.5.7 Discussion: Neutrophils and Eosinophilic Granulocytes, Lymphocytes

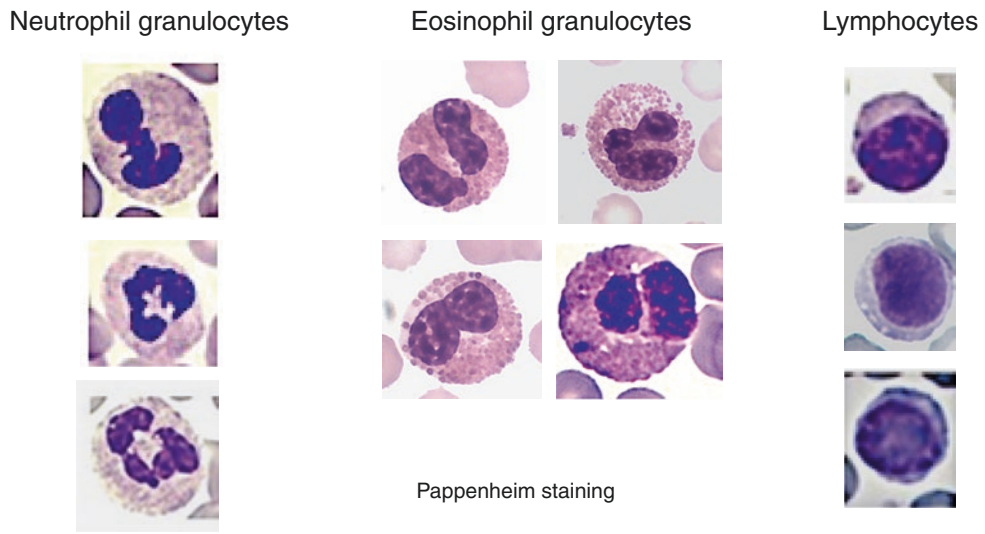


Fig. 11.25 In order to better differentiate the various species, the leukocytes are visualized in Pappenheim-stained blood smear samples. Generally, neutrophil granulocytes multiply in urine due to inflammatory reactions in the kidneys and urinary tract. In the case of allergic reactions, eosinophilic granulocytes with a large vesicular granule and a bi-segmented nucleus may also be excreted. This type of leukocyte cannot be differentiated in urine without special staining (Hansel's stain). Renal transplant rejection reactions may be accompanied by lymphocyturia. It is essential not to confuse lymphocytes with renal epithelial cells due to the round nucleus

11.5.8 Histiocytes (Macrophages)

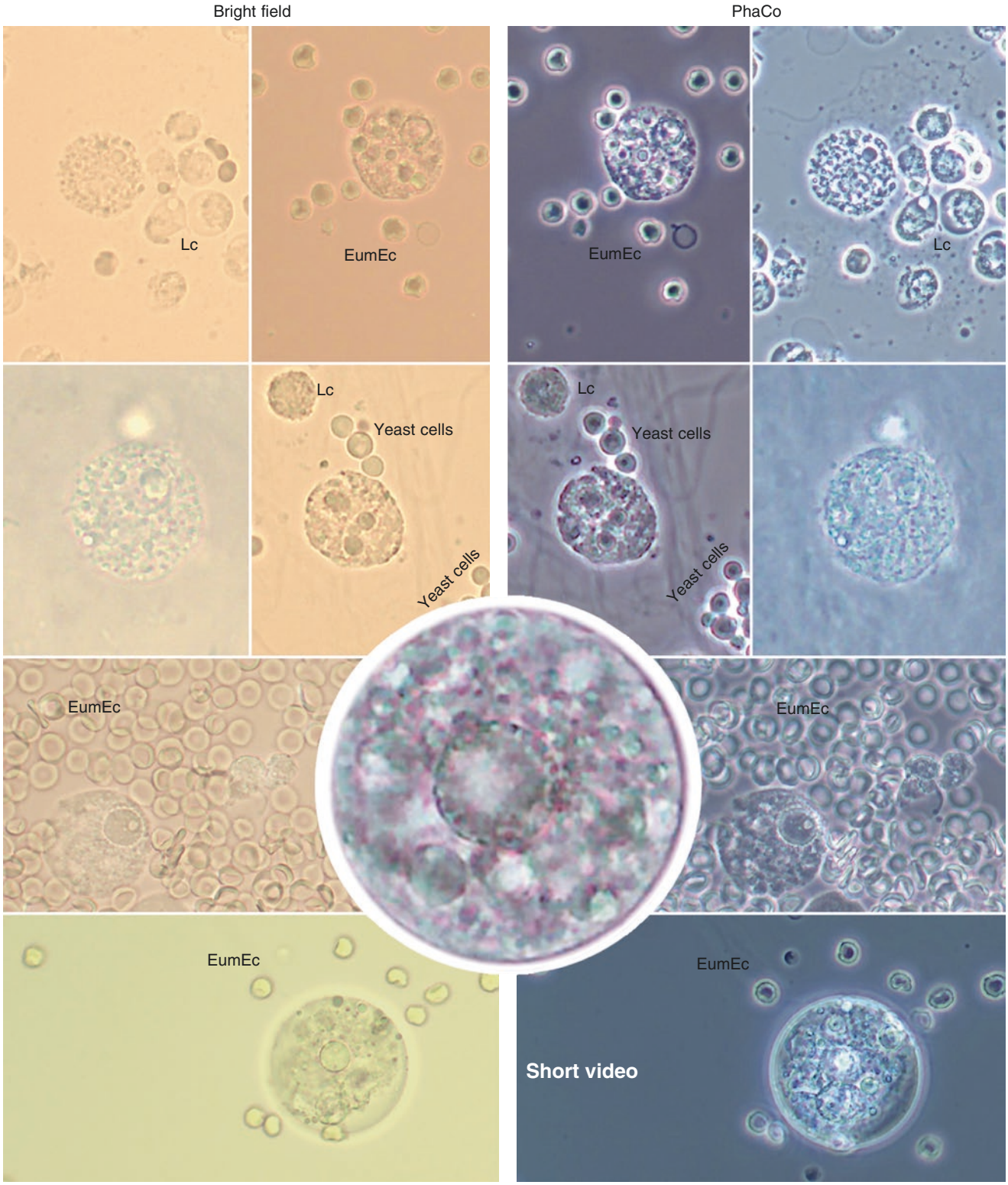


Fig. 11.26 Histiocytes (macrophages). Image magnification: histiocyte with clearly visible evenly distributed granules, vacuoles, and phagocytized constituents. (see Video 11.5)

11.5.9 Old Histiocytes (Macrophages)

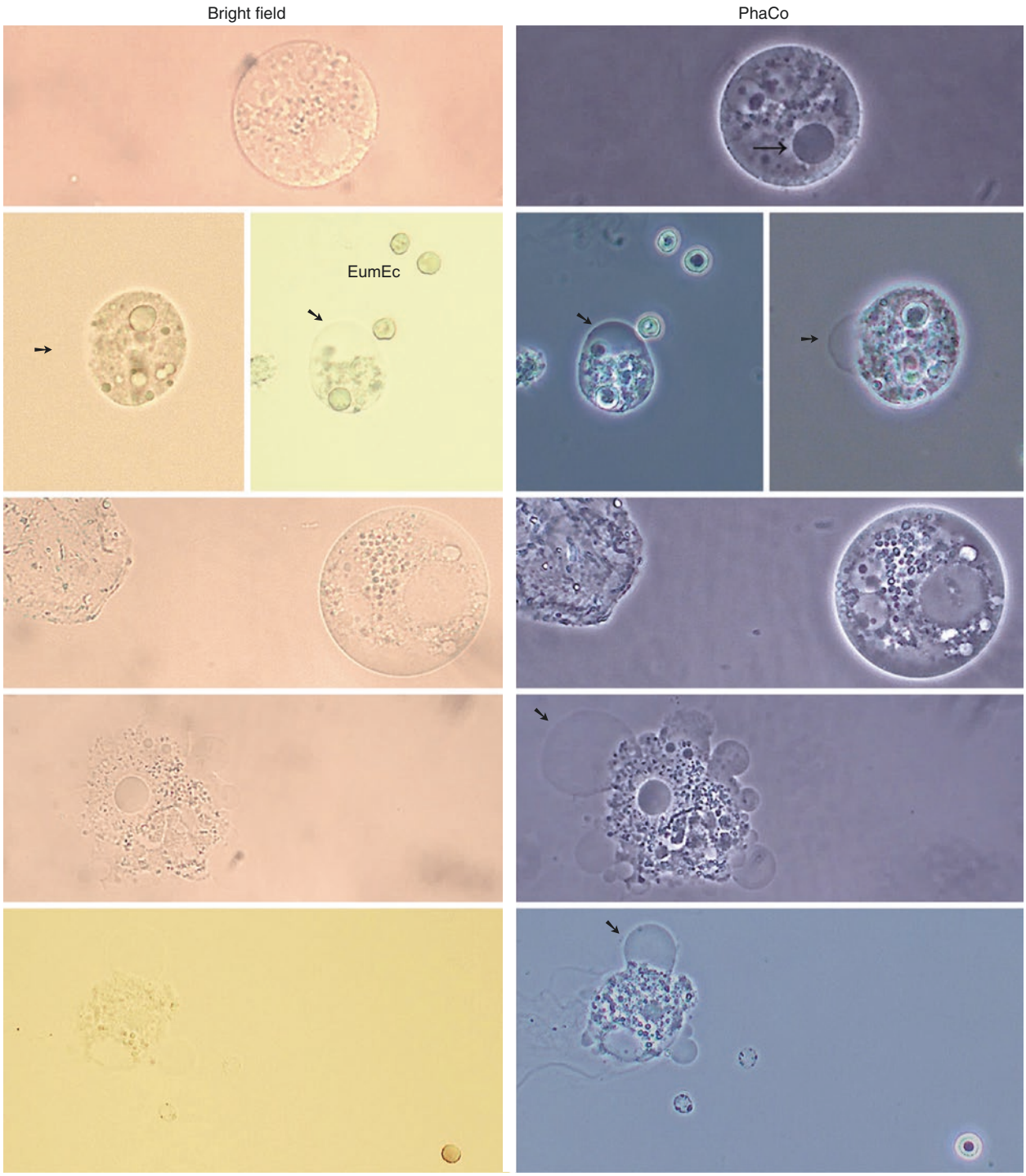


Fig. 11.27 Histiocyte aging criteria: strikingly dark nucleus (→) with altered nuclear chromatin. The aging-related vesicles are clearly visualized in PhaCo (➙)

11.6 Parasites

11.6.1 Trichomonads

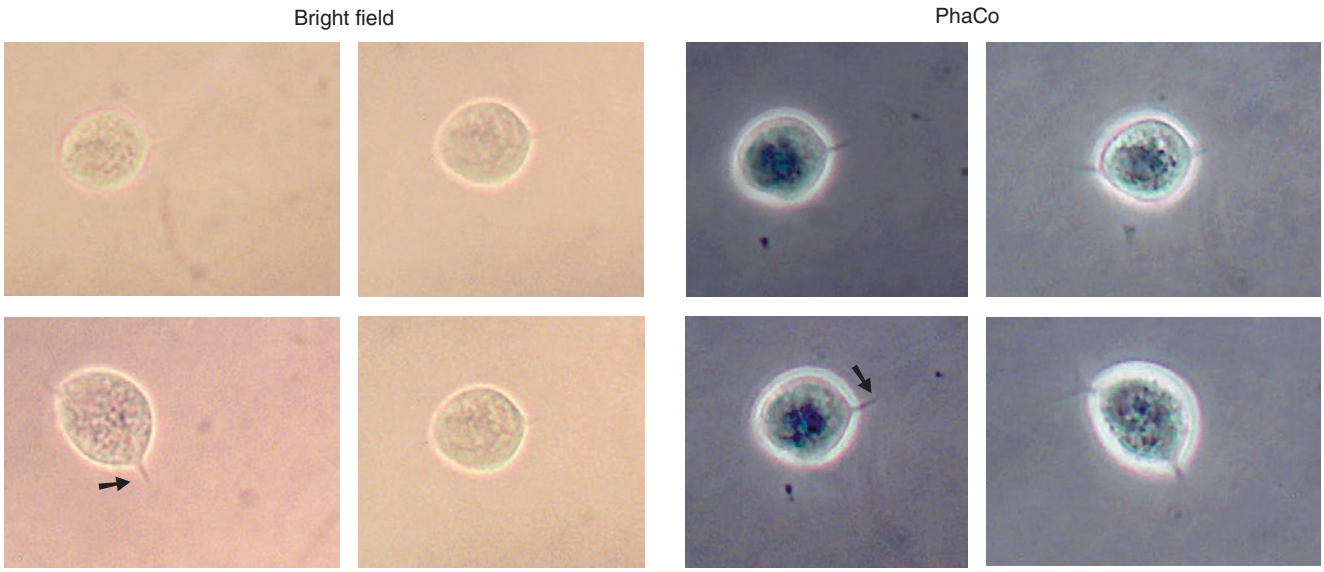


Fig. 11.28 Since trichomonads in fresh urine rapidly migrate across the visual field due to their mobility (see flagellates (➞)), they can be reliably distinguished from leukocytes. Due to this mobility, differentiation is unequivocal, since motionless trichomonads are barely distinguishable from leukocytes

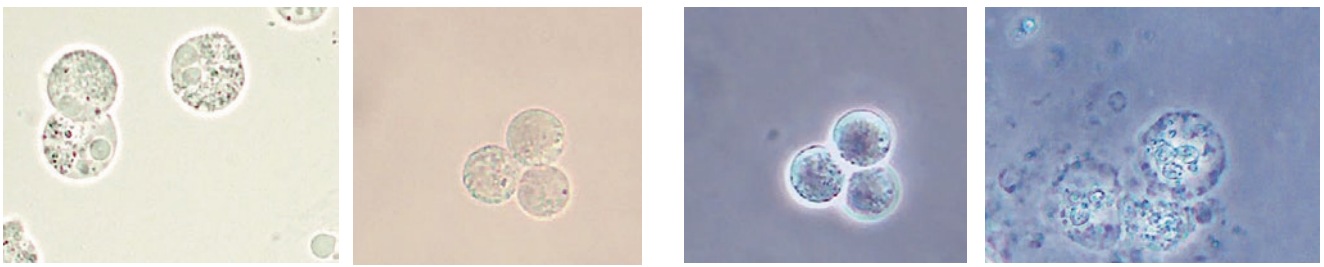


Fig. 11.29 For comparison: large- and small-cell leukocytes

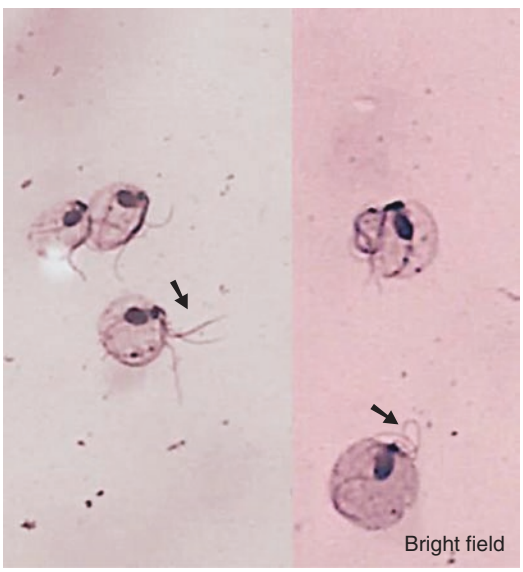


Fig. 11.30 Stained trichomonads (Giemsa stain). In stained specimens, the trichomonad flagellates (➞) can be clearly visualized and compared with Fig. 11.28 of the unstained native specimens in bright-field and PhaCo modes

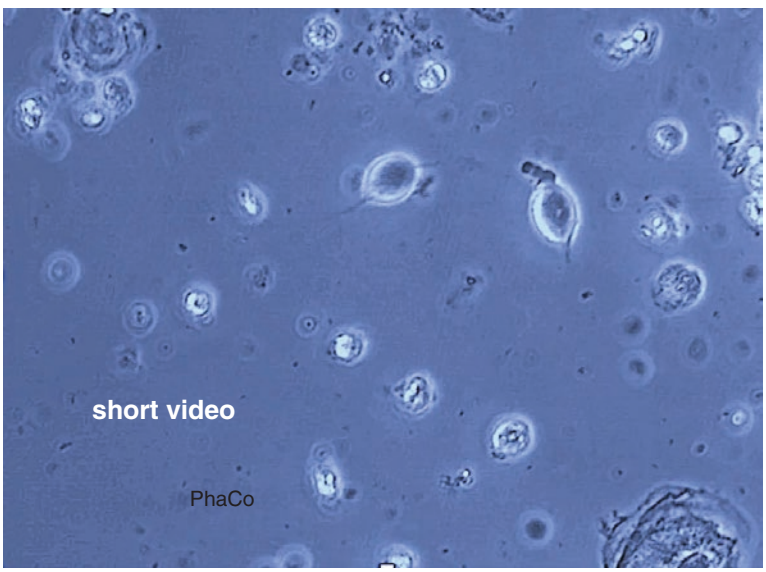


Fig. 11.31 The trichomonads in the short film can be easily distinguished from the surrounding leukocytes due to their mobility. (Courtesy of the Nephrology Laboratory, Medical Department I and Outpatient Department, University Medicine Mainz, Germany). (see Video 11.6)

11.6.2 *Schistosoma haematobium* Eggs

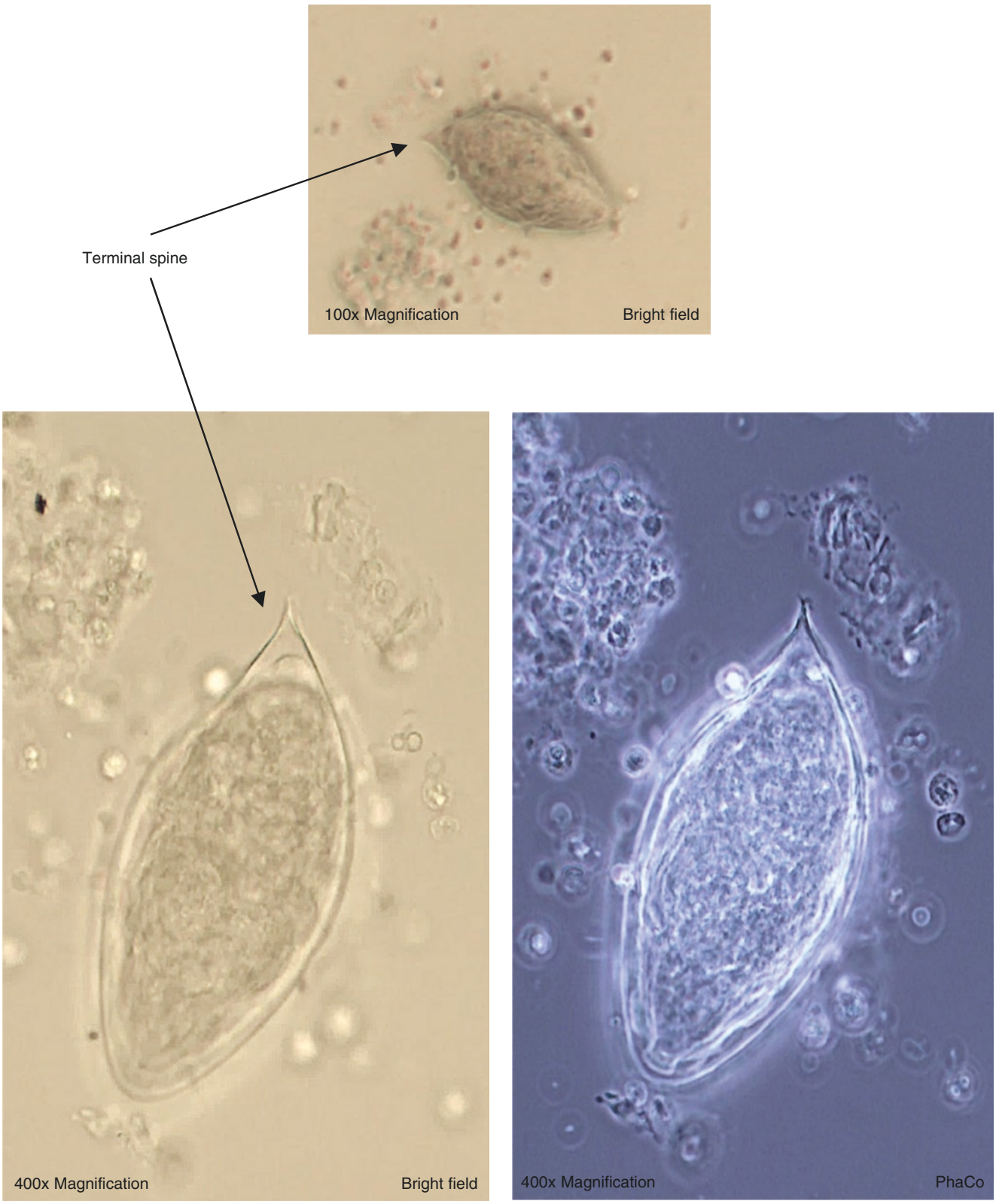


Fig. 11.32 *Schistosoma haematobium* egg I: these eggs are extremely large and easily seen even at 100× magnification. Eumorphic erythrocytes, leukocytes, and transitional epithelial cells lie next to the egg. Due to the enormous thickness of the egg, not all surrounding cells can be sharply visualized

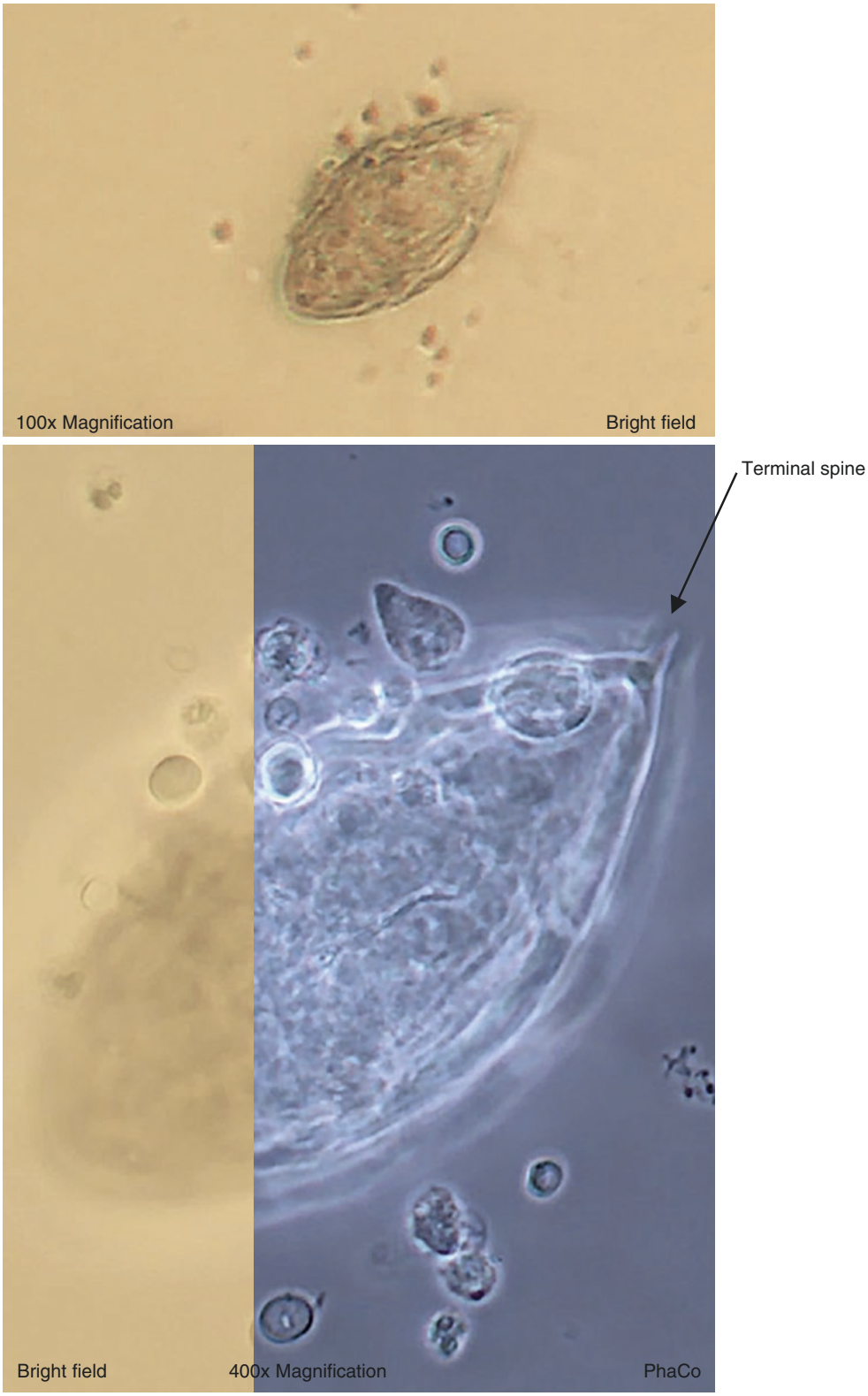
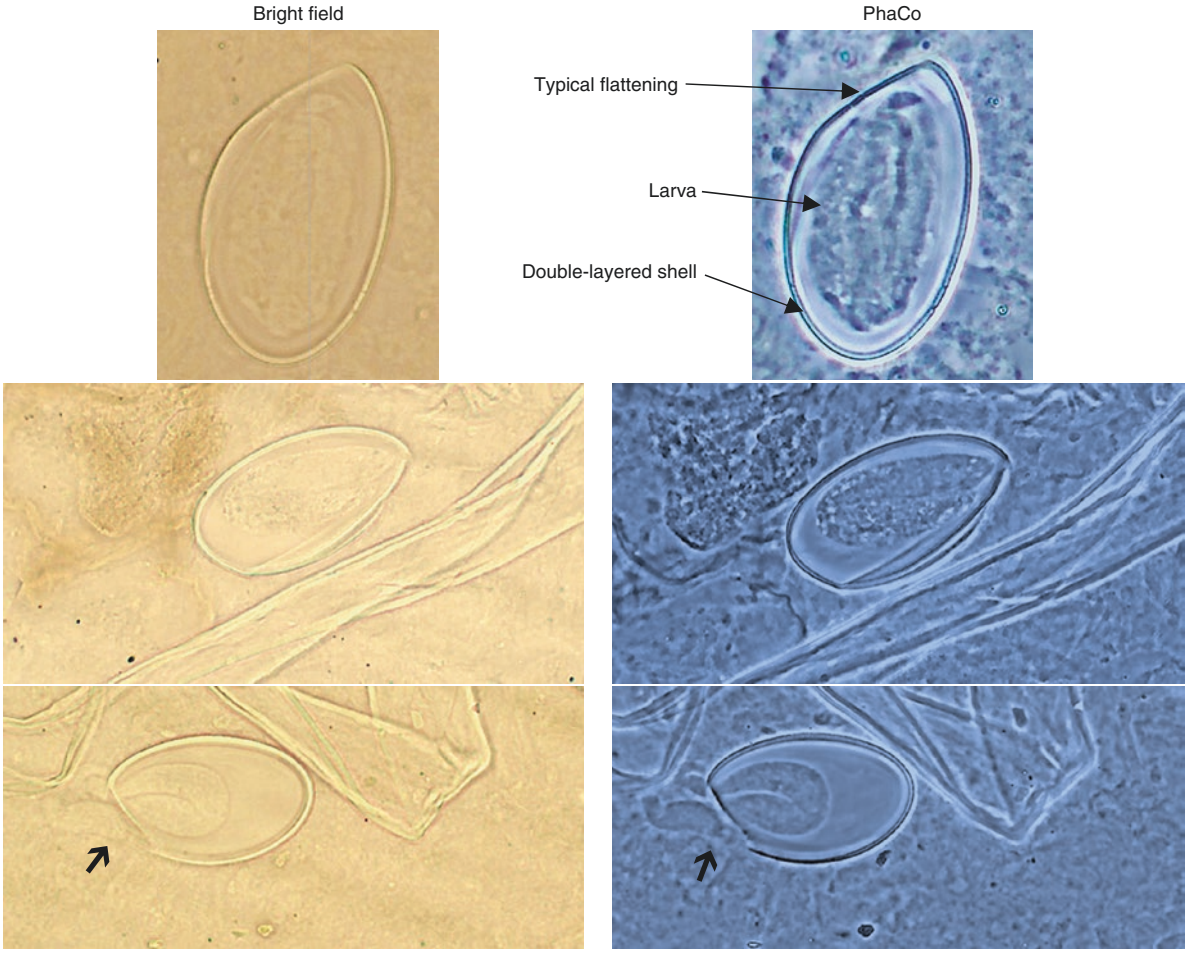
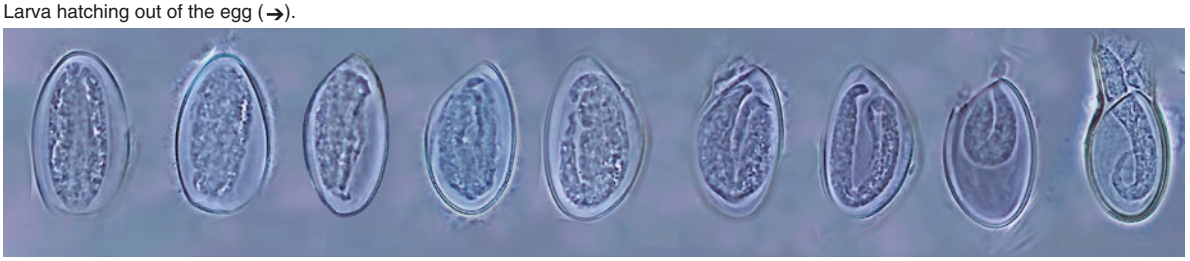


Fig. 11.33 *Schistosoma haematobium* egg II: at 100× and 400× magnification

11.6.3 *Enterobius vermicularis* Eggs



Larva hatching out of the egg (➔).



Developmental stages of the larva up to hatching.

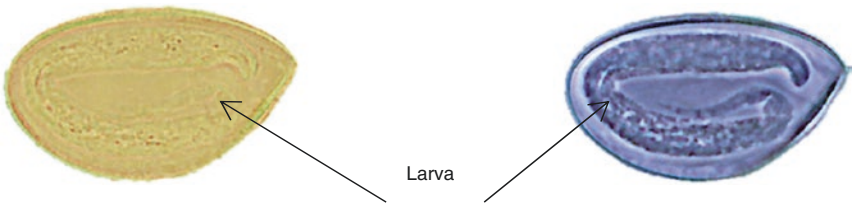


Image magnifications: the larvae in the eggs are easily visualized in phase-contrast mode.

Fig. 11.34 *Enterobius vermicularis* eggs

11.7 Epithelial Cells: An Overview

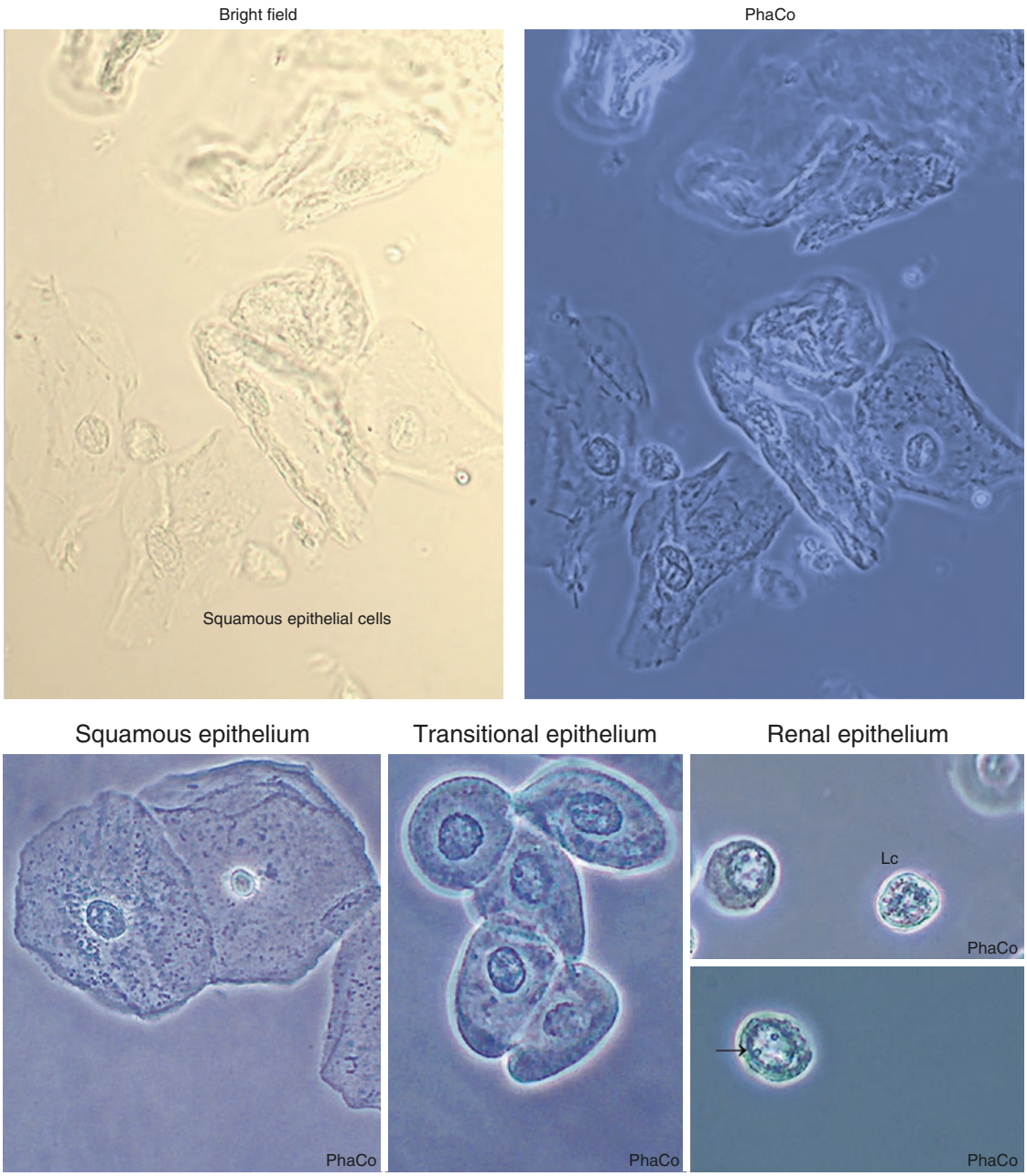


Fig. 11.35 The difficult distinction between a renal epithelial cell and a transitional epithelial cell is made at least slightly easier if one takes into account the fact that a renal epithelial cell is marginally larger than a leukocyte. The differentiation of small fat droplets (→) in the cytoplasm of the renal epithelial cell is also helpful, since only these epithelial cells are able to absorb fat particles

11.7.1 Squamous Epithelial Cells

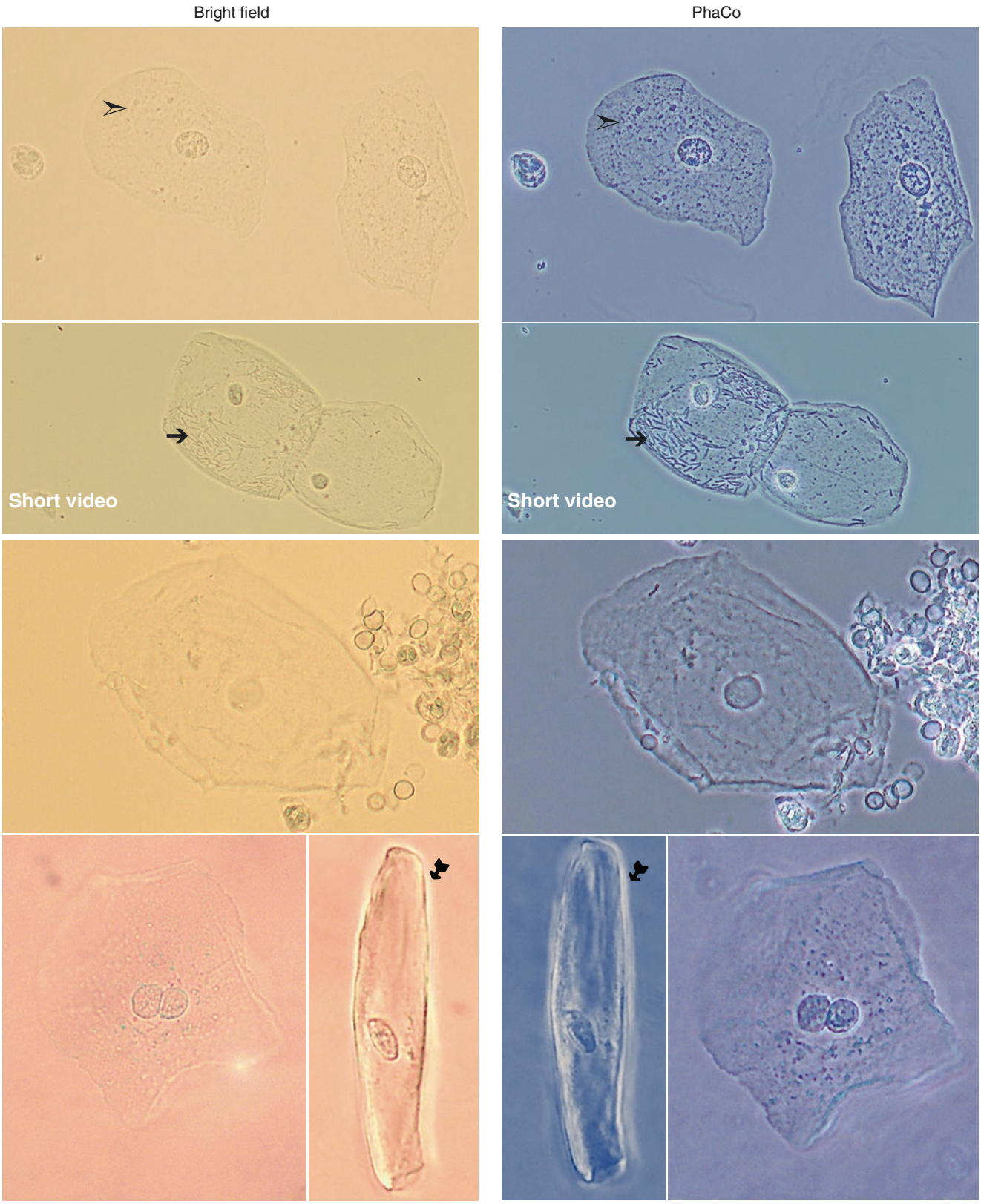


Fig. 11.36 Bacteria (➔) as well as crystals become easily entangled in the squamous epithelial cells. The cytoplasm sometimes appears transparent and granulated (➢). Squamous epithelial cells can appear extremely narrow from the side view (➳) and, if no nucleus is visible, simulate a cast. (see Videos 11.7 and 11.8)

11.7.2 Squamous Epithelial Cells: Cell Groups

If more than seven to eight squamous epithelial cells are counted per visual field, one can assume that the urine sample was not taken from midstream urine.

Epithelial cells are virtually invisible in bright-field mode. Small, brightly luminescent fat droplets (➢) lie at the edge of the squamous epithelial cell's nucleus.

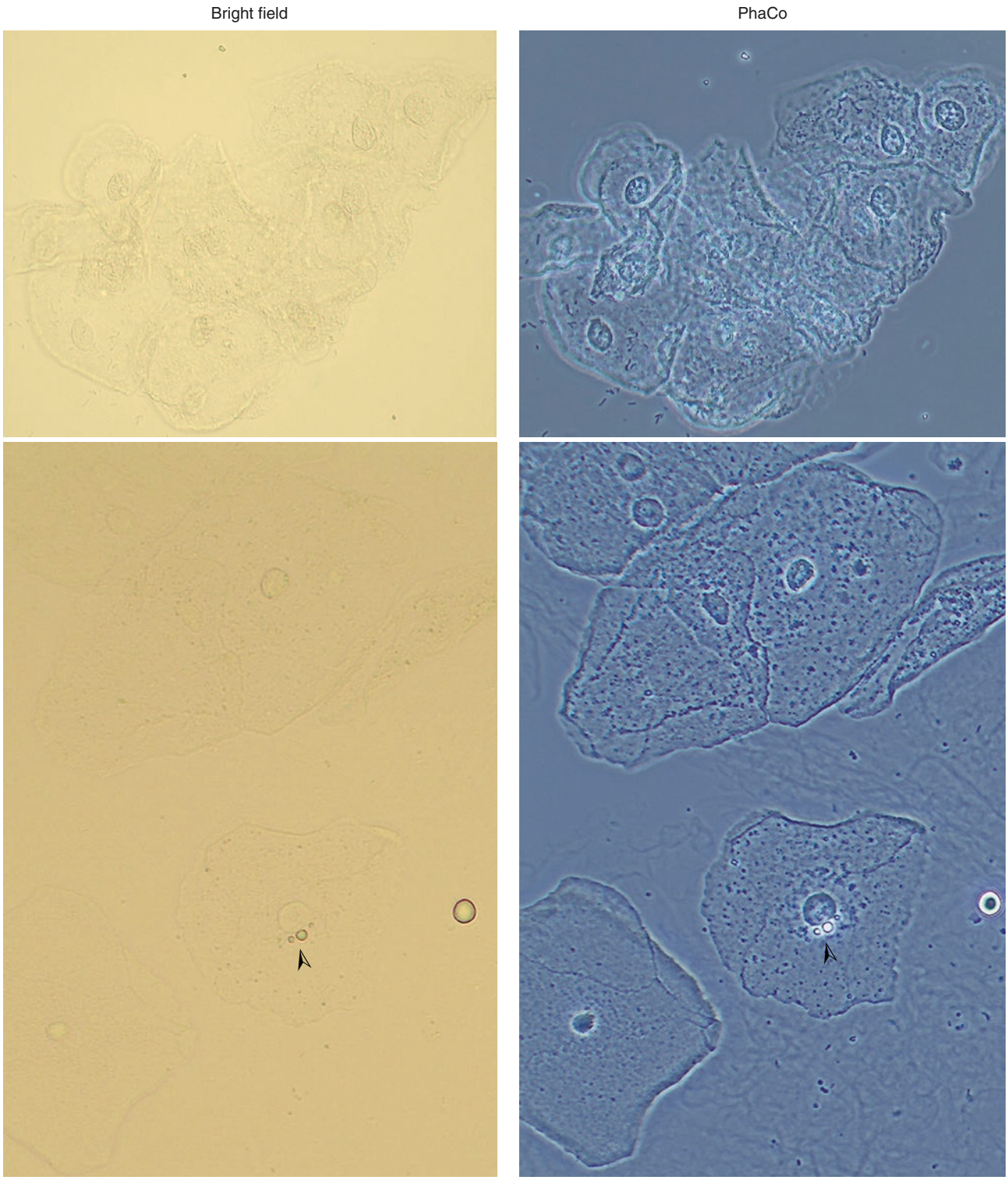


Fig. 11.37 Squamous epithelial cells: cell groups

11.7.3 Transitional Epithelial Cells (Urothelium)

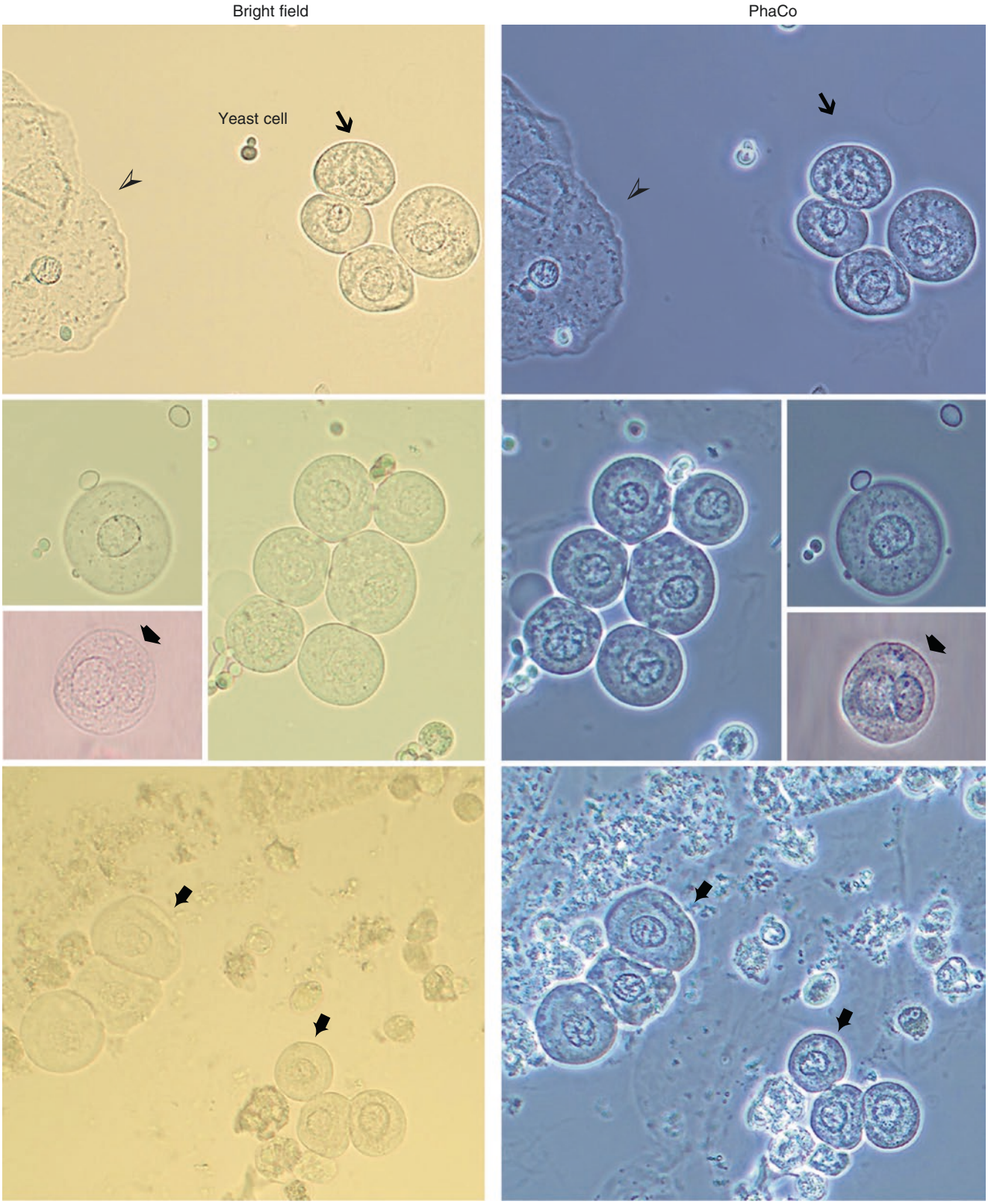


Fig. 11.38 Transitional epithelial cells (urothelium), round/oval (➔), are easily distinguished from the squamous epithelial cells (➢). Multinucleation (◗) is normal and not a sign of malignancy [Rathert et al., *Urinzytologie und Sedimentanalyse*, Springer 2018 (5), p. 31ff]. Approximately 19% of urothelial umbrella cells have two nuclei. Urothelial cells vary greatly in size (➨)

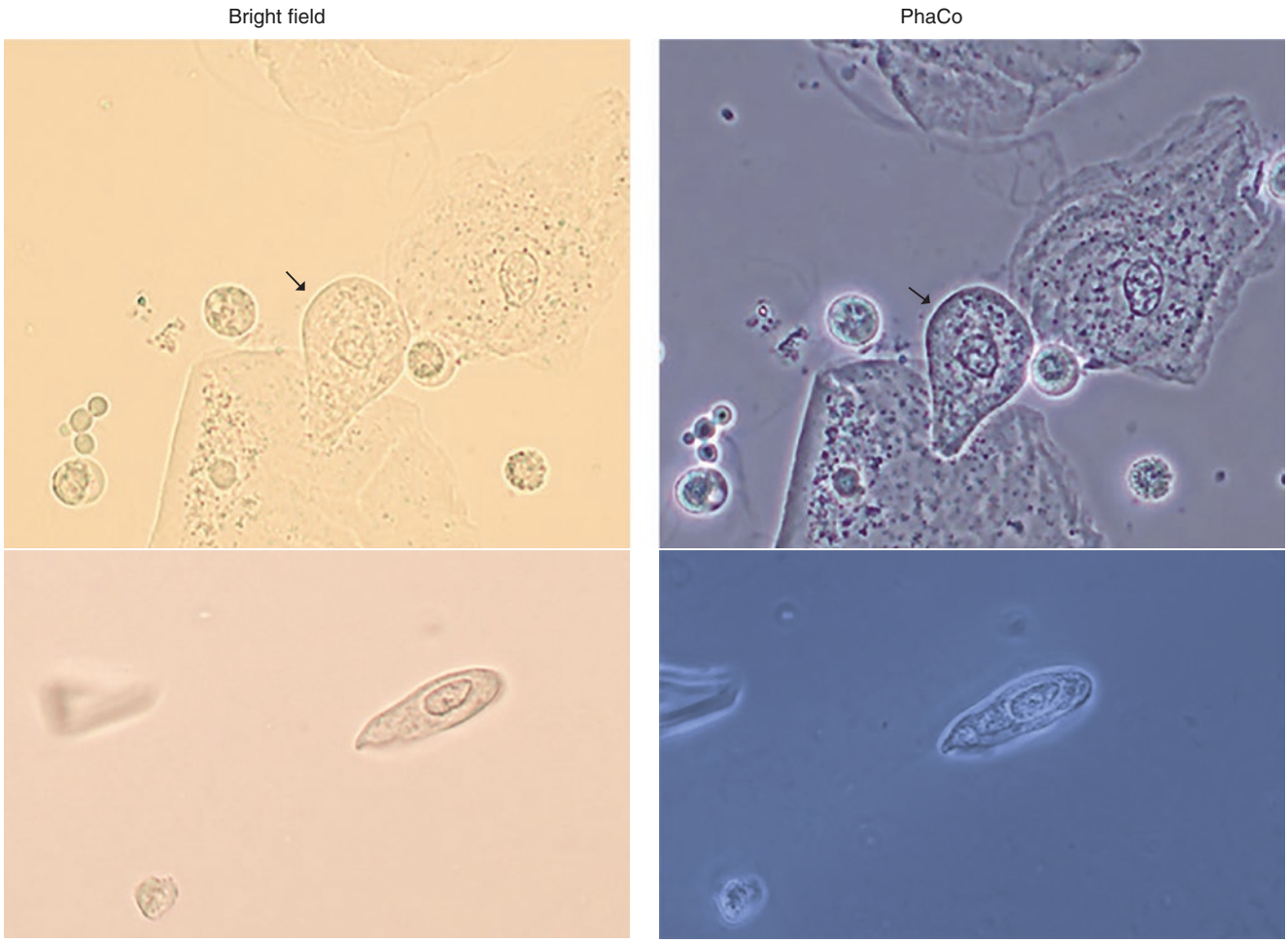


Fig. 11.39 Transitional epithelial cells (urothelium), with tail-like projections. A transitional epithelial cell (→) lies between two squamous epithelial cells, leukocytes, and yeast cells

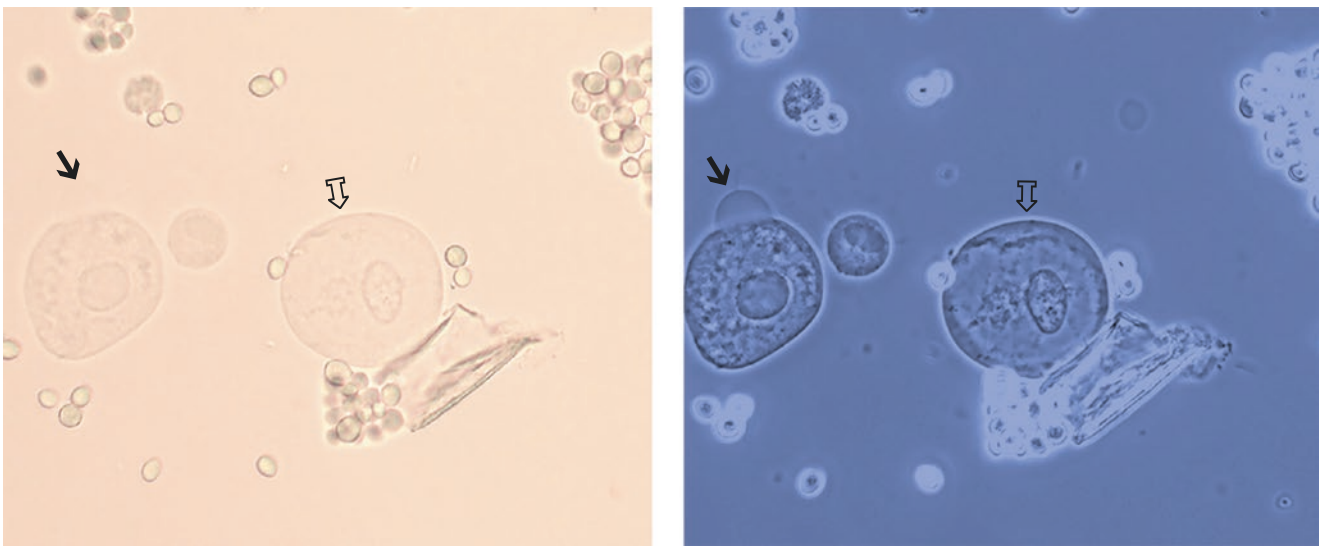


Fig. 11.40 Old transitional epithelial cells (urothelium). Cell-aging processes alter cell morphology. Aging-related vesicles attached to the cell contour (➔) and the altered cell border (⇨) can be seen

11.7.4 Deep Urothelial Cells

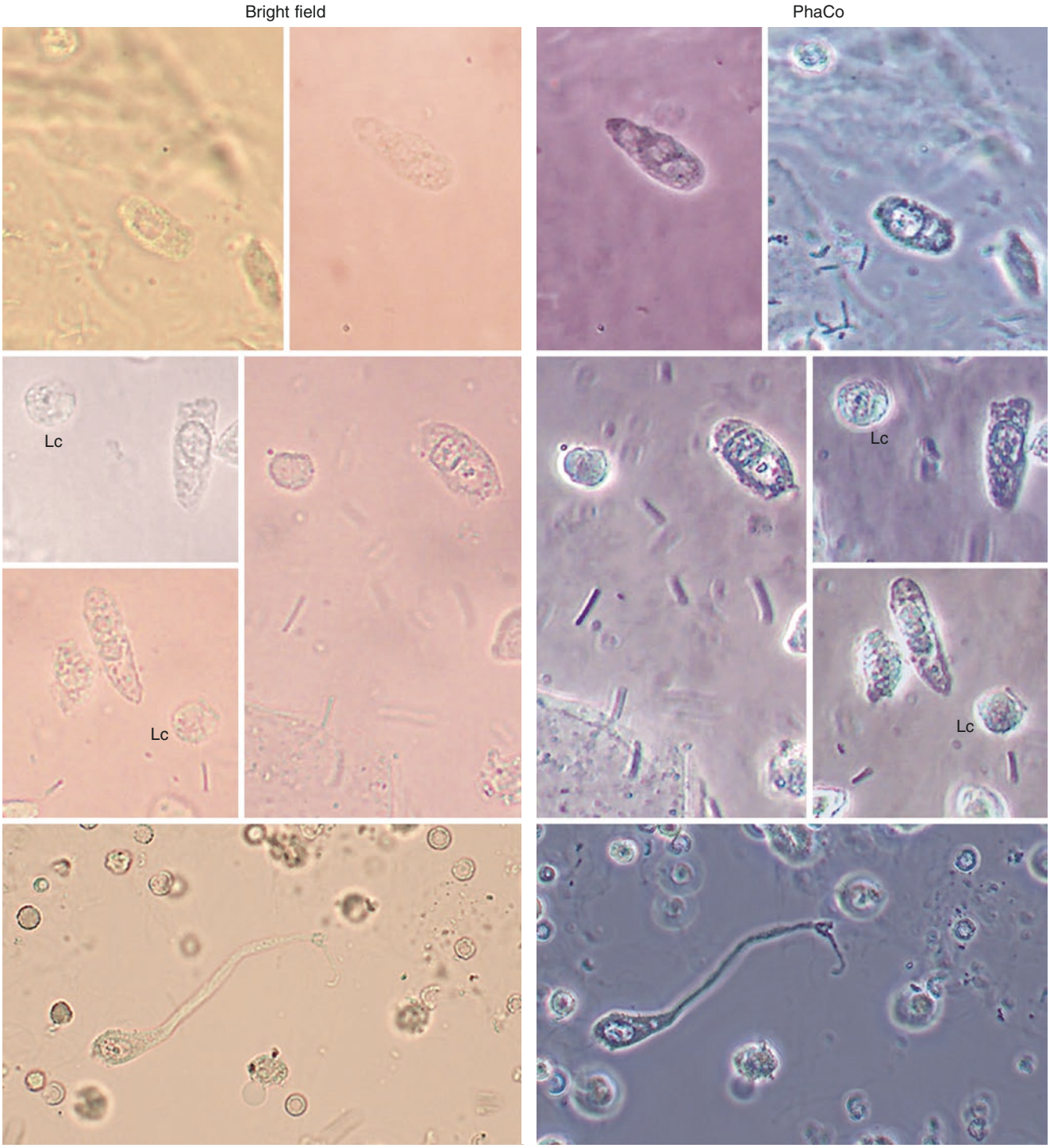


Fig. 11.41 Deep urothelial cells: cubic and with tail-like projections

11.7.5 Comparison: Transitional Epithelial Cells–Old Leukocytes

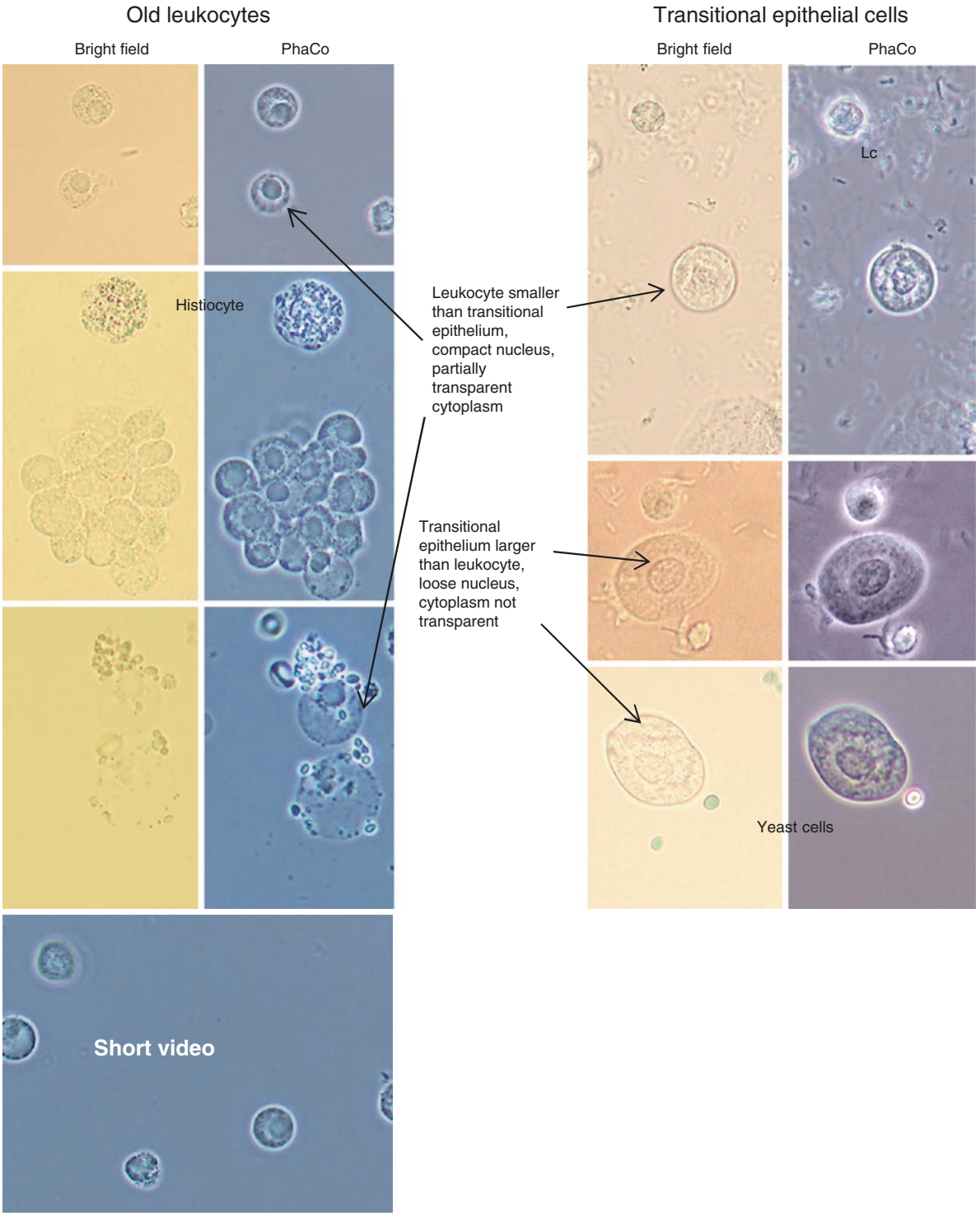
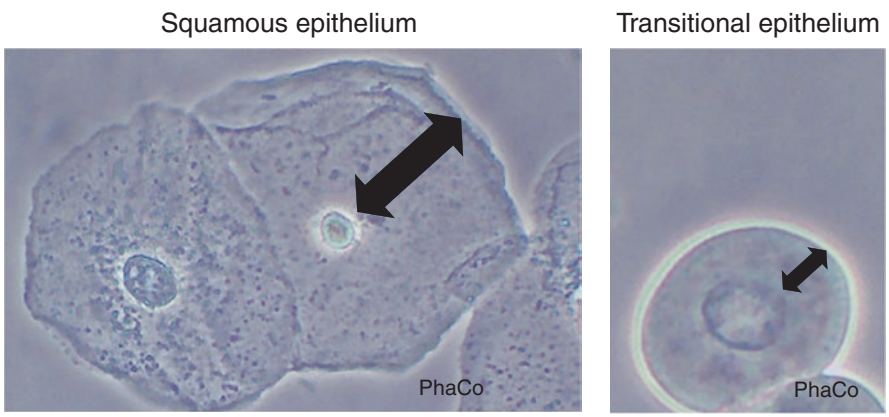


Fig. 11.42 Comparison: old leukocytes–transitional epithelial cells. The similar nucleus–cytoplasm ratio of the two cell types causes problems in differentiation. The short video shows old, enlarged leukocytes with a round nucleus and granular motility (Sternheimer-Malbin cells). (see Video 11.9)

11.7.6 Comparison: Squamous Epithelium–Transitional Epithelium



Large epithelium with angular border, small pyknotic nucleus (compact, dark) or somewhat larger with loose chromatin, each with perinuclear zones of brightening, i.e., halo around the nucleus, nucleus–plasma ratio shifted in favor of the cytoplasm

Smaller than squamous epithelium, round cell, nucleus appears to be transparent, loose chromatin, nucleus–plasma ratio shifted in favor of the nucleus

Fig. 11.43 Comparison: squamous epithelium–transitional epithelium

11.7.7 Renal Epithelial Cells (Renal Tubular Epithelial Cells)

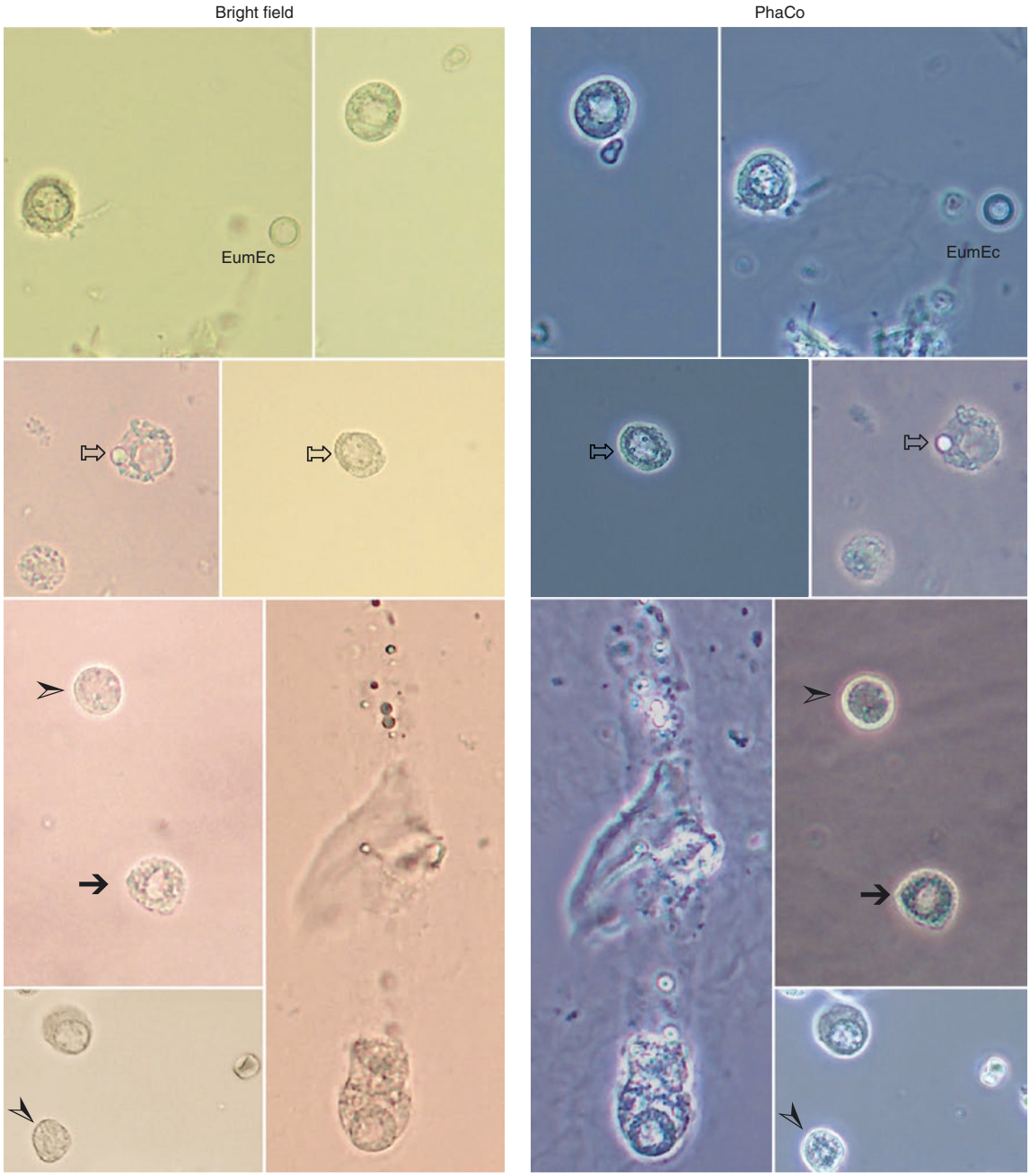


Fig. 11.44 Renal epithelial cells are the smallest epithelial cells excreted in urine. Important morphological criteria: a renal epithelial cell (➔) is somewhat larger than a leukocyte (➢) and always has a round, compact nucleus. Storage of fat droplets (⇨) facilitates differentiation. Renal epithelial cells must not be confused with small urothelial cells or deep urothelial cells!

11.7.8 Old Epithelial Cells

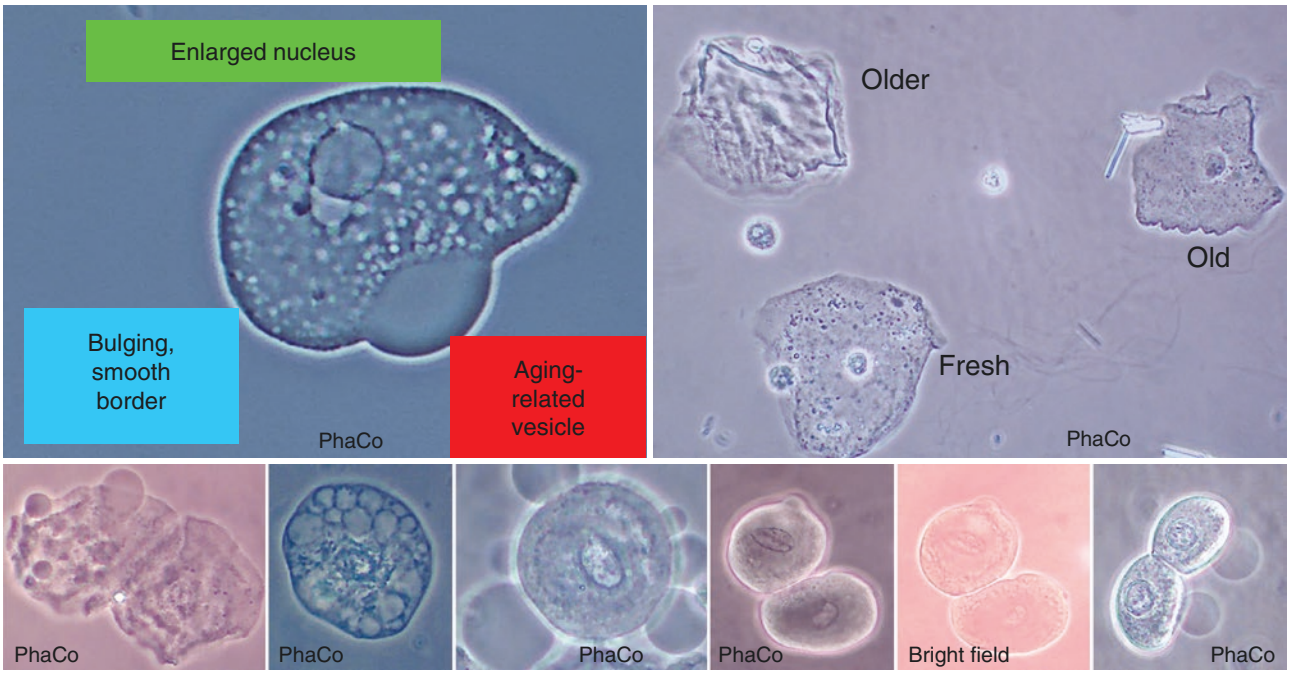
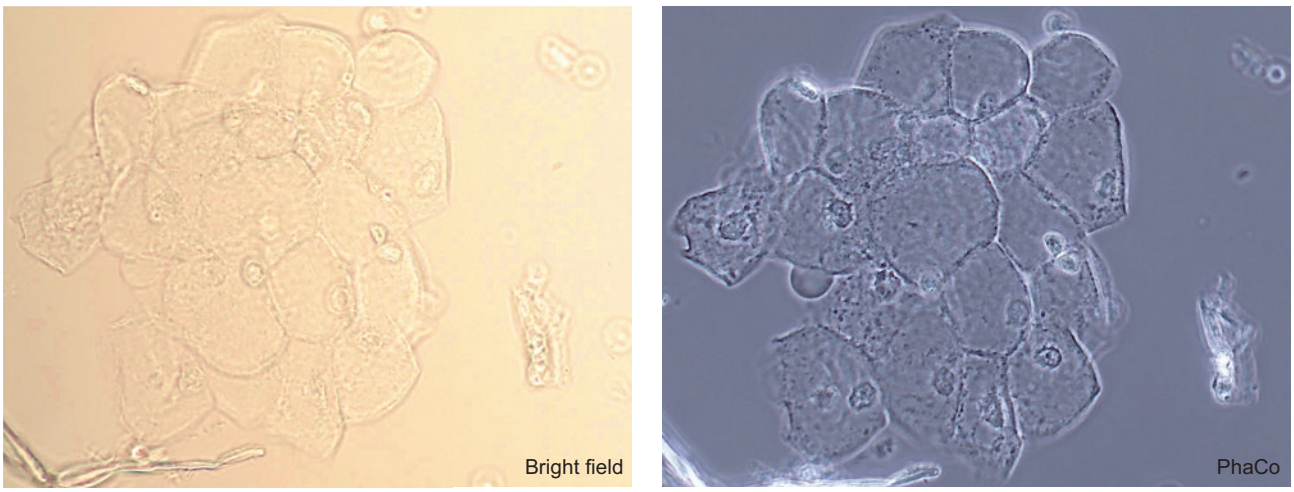
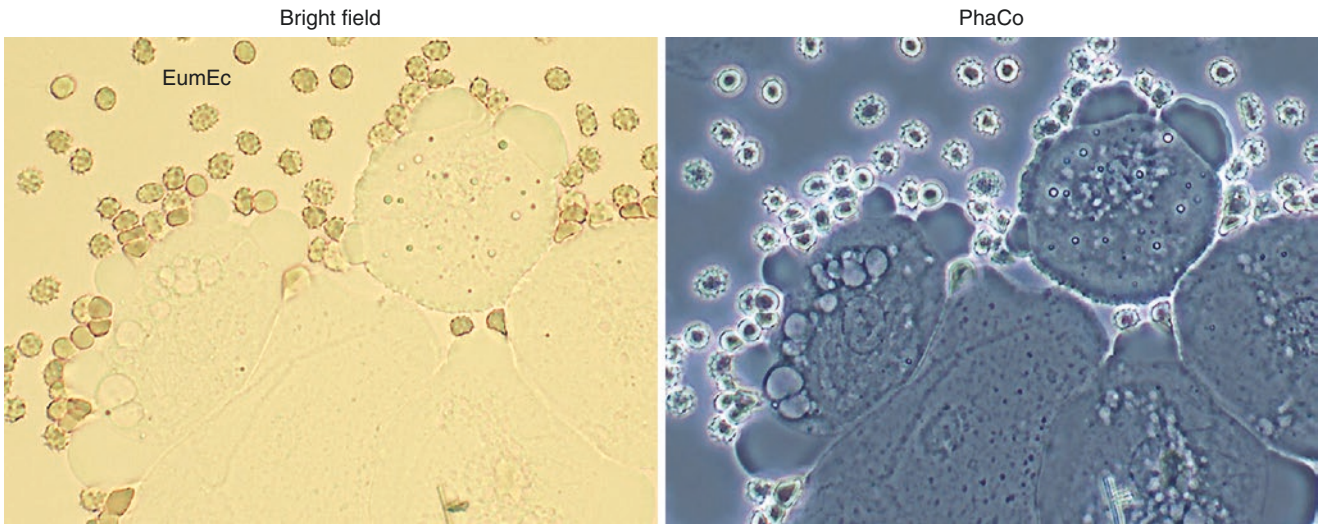


Fig. 11.45 Old squamous epithelial cells can assume a round shape, making them easy to confuse with transitional epithelial cells. Epithelial cells in catheter urine that has been stored for a prolonged period in urine bags can change their appearance to such an extent that they are easily confused with atypical epithelial cells

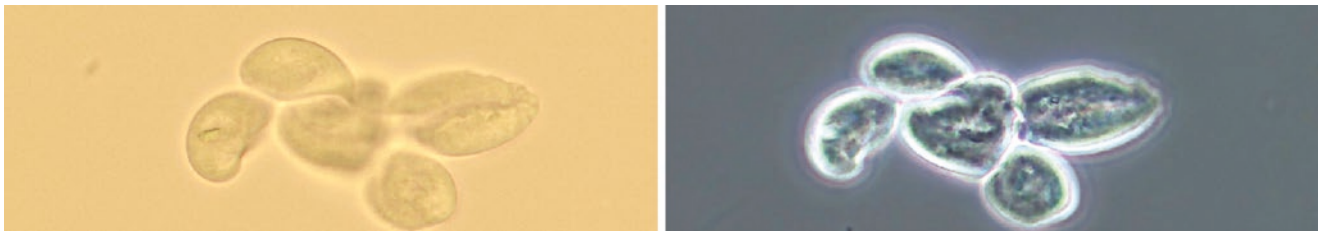


The nucleus in the individual epithelial cells is sometimes no longer visible.

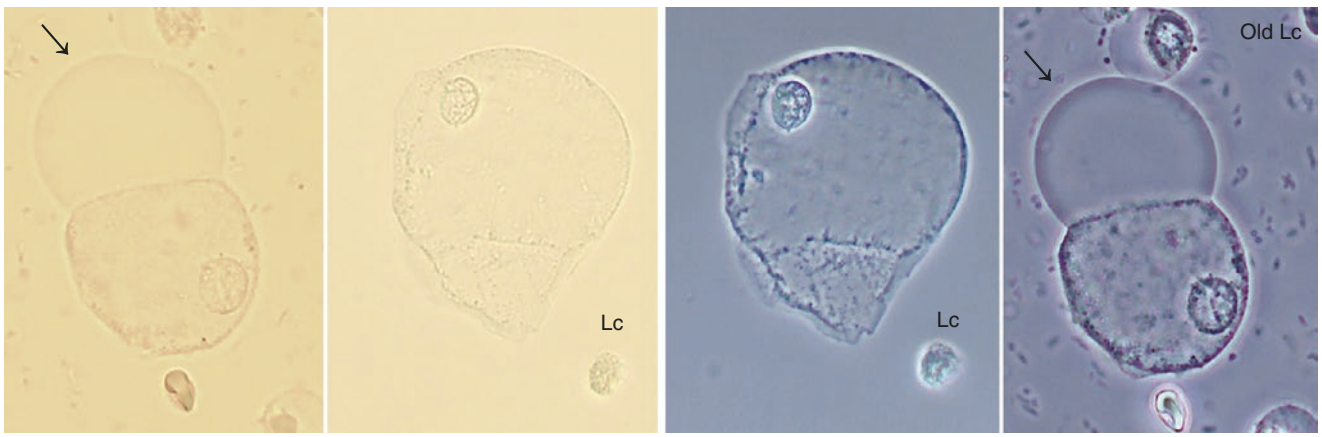
Fig. 11.46 Old epithelial cells: cell accumulation



Numerous squamous epithelial cells with aging-related vesicles and vacuolized cytoplasm lie next to eumorphic (thorn apple-shaped) erythrocytes.



Only on the basis of the cell contour can it be assumed that this is an epithelium. Precise classification of the epithelium is no longer possible, since the cell borders are poorly defined and the cell nuclei are not visible.
One can assume that the urine sample is old (>2h) if predominantly obsolete epithelial cells are identified.



Huge aging-related vesicles (→) can form on the epithelium. Using unclean working materials (slides and cover glasses) can also cause morphological alterations to cells.

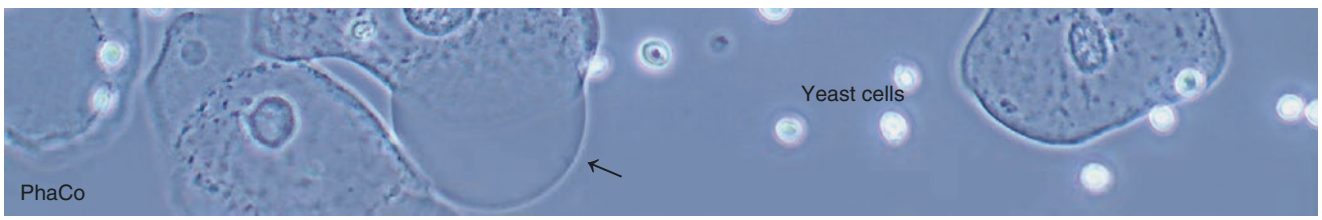


Fig. 11.47 Old epithelial cells

11.7.9 Oval Fat Bodies–Intracellular Lipid Droplets

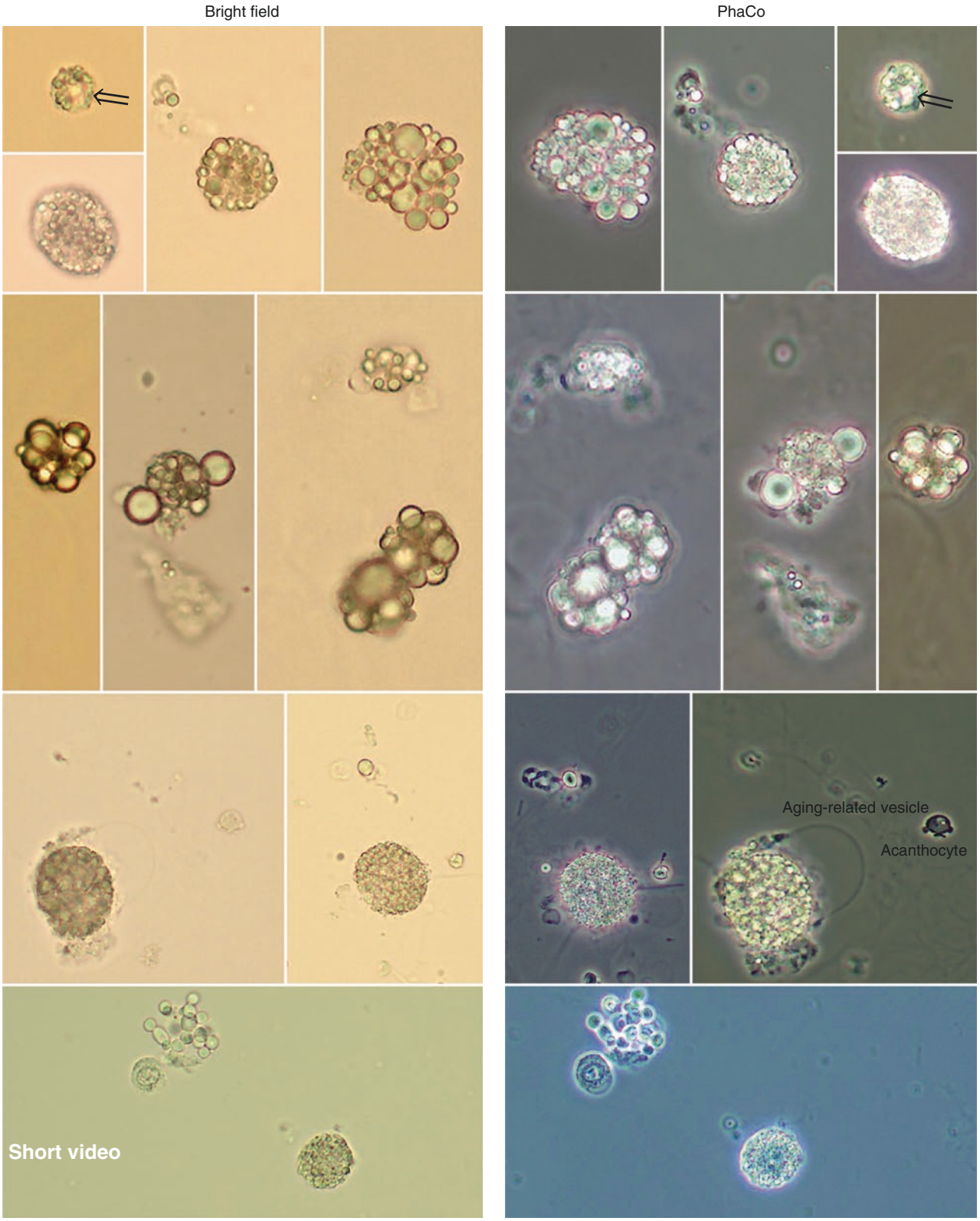


Fig. 11.48 The fat droplets light up bright yellow in both bright-field and phase-contrast mode. If the fine-focusing knob of the micrometer knob is constantly operated, the fat appears shiny. Since a renal epithelial cell can have varying densities and be packed with fat particles of varying size, an oval fat body can reach a considerable volume. However, if the cell is not densely packed with fat, the nucleus (⟹) can still be seen. (see Video 11.10)

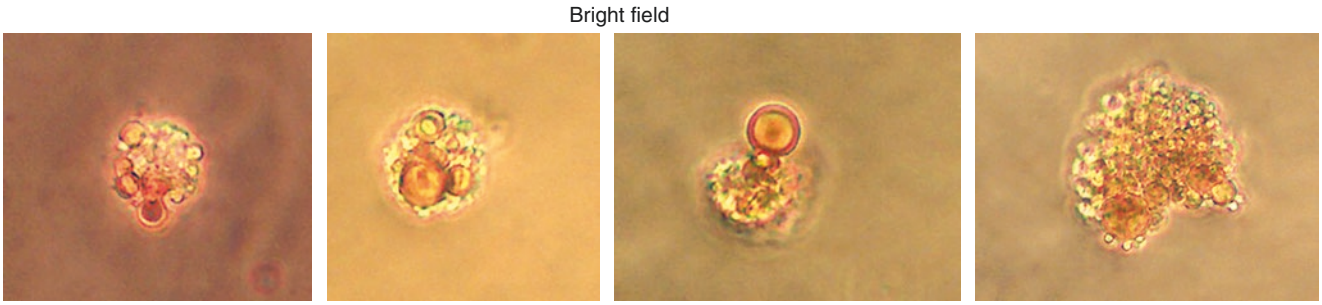
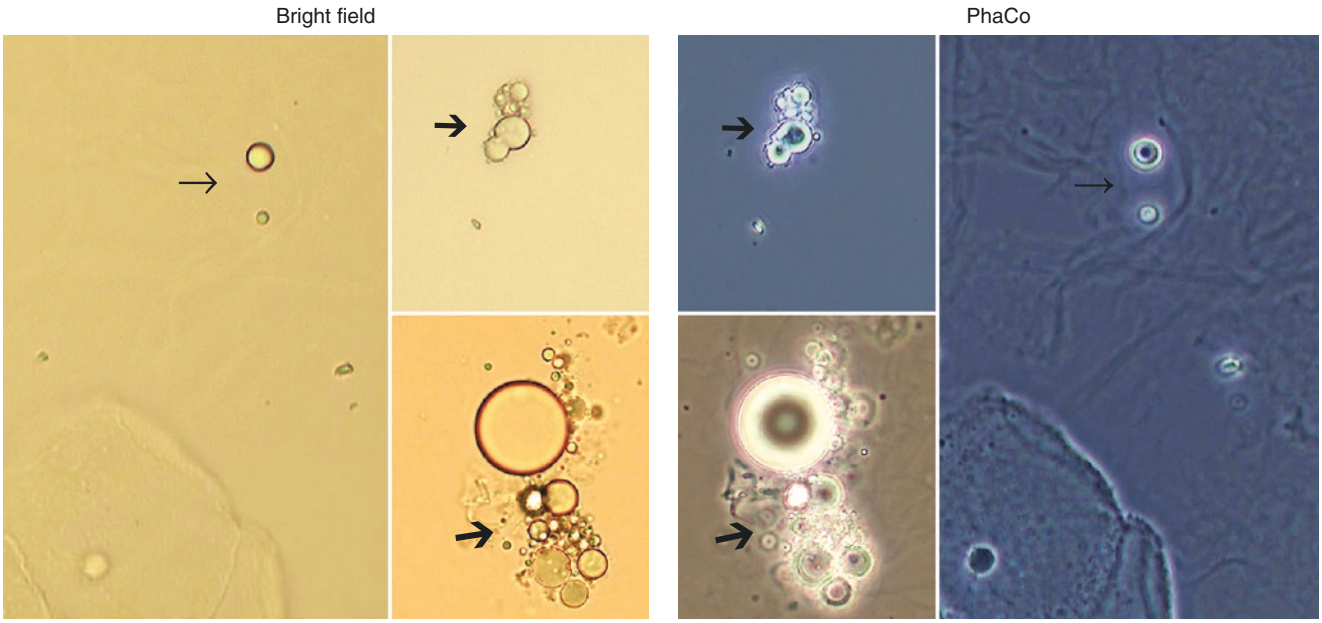
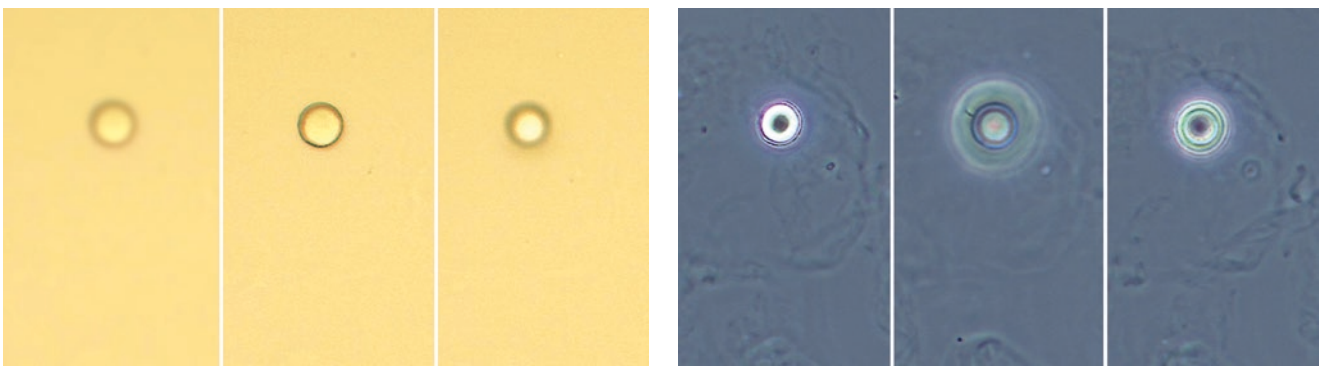


Fig. 11.49 Oval fat bodies stained with Sudan IV

11.7.10 Discussion: Extracellular Lipid Droplets



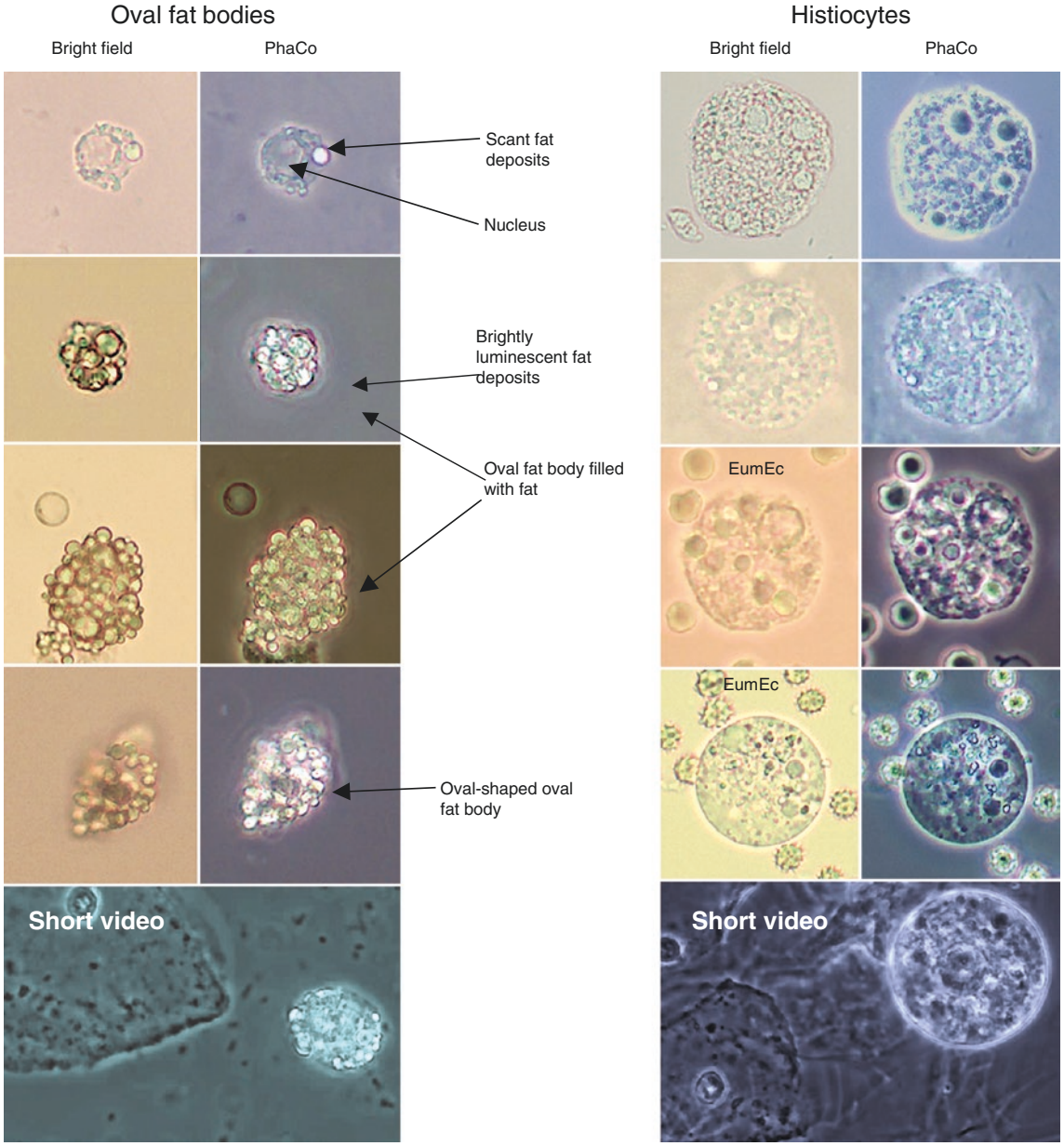
In these images, lipid particles lie extracellularly, alone (→), or in a cluster (➔) in small as well as larger drops. Free-floating lipid particles can be due to disease, e.g., nephrotic syndrome, or may also occur as artifacts due to the use of ointments or suppositories.



By using the fine-focusing of the micrometer knob, fat droplets (in contrast to an erythrocyte) light up with varying degrees of brightness and shininess.

Fig. 11.50 Extracellular lipid droplets

11.7.11 Comparison: Oval Fat Bodies–Histiocytes



Oval fat bodies with fat droplets in a comparatively *inhomogeneous* or *grape-like* arrangement, covering the contour of the renal epithelium and thereby making the cell border invisible.

Histiocytes have a more or less *even* distribution of granules, vacuoles, and phagocytized components that do not cover the cell contour. These cells are usually larger than oval fat bodies.

Fig. 11.51 Comparison: oval fat bodies–histiocytes. (see Videos 11.11 and 11.12)

11.7.12 Comparison of Oval Fat Bodies–Histiocyte–Leukocyte with Phagocytized Yeast Cells–Old Epithelial Cells

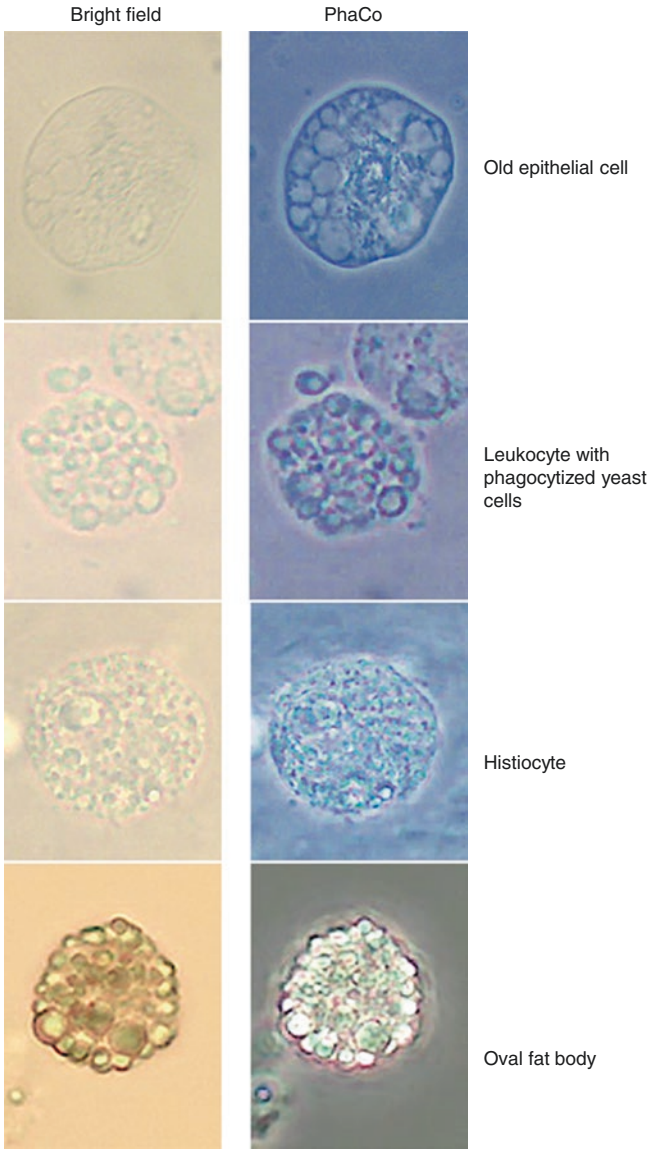


Fig. 11.52 The view sizes of the individual cells were deliberately adjusted in these images! However, this makes it possible to visualize and compare the interior of the cell structures

11.7.13 Decoy Cells

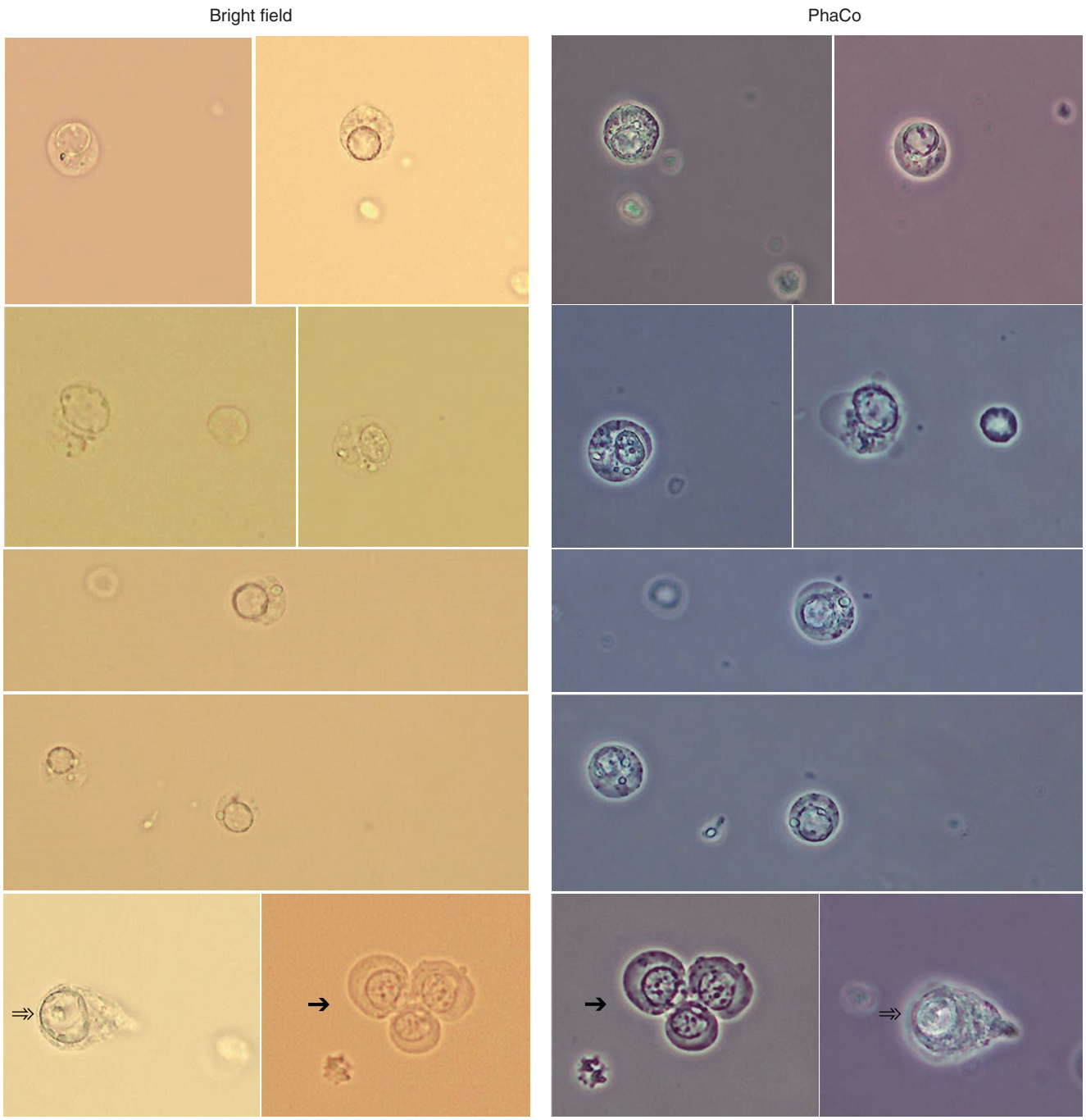


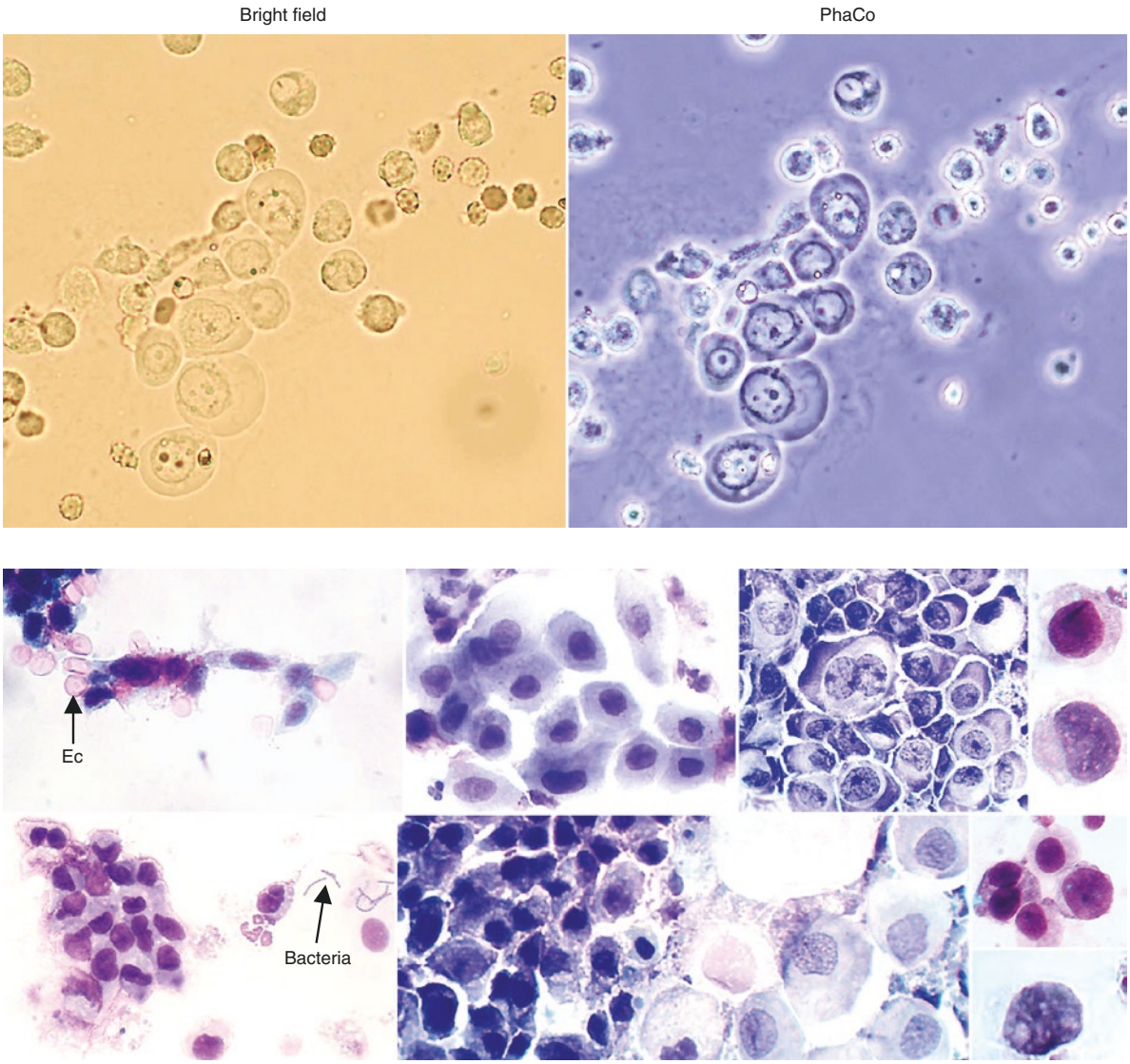
Fig. 11.53 Characteristic morphology of decoy cells: enlarged nucleus, altered nuclear chromatin (ground glass-like), partially displaced to the border of the nucleus and densified. Both renal epithelial cells (➔) and transitional epithelial cells (⇛) are subject to these morphological changes. Decoy cells need to be distinguished from atypical cells/tumor cells and obsolete cells. Therefore, one can express only a suspicion of decoy cells in the findings of the microscopic native specimen

11.7.14 Tumor Cells

The presence of atypically shaped epithelial cells in the unstained native specimen can have various causes. Therefore, one can only speak of "suspected atypical cells" in the findings. Further urine cytological investigation is required.

Morphological criteria of atypical cells*
– Enlarged, multiform cell nucleus, shift in nucleus–plasma ratio
– Altered nuclear membrane
– Chromatin propagation and chromatin coarseness
– Enlargement and propagation, etc., of nucleoli

*See Rathert et al.: *Urinzytologie und Sedimentanalyse*, Springer 2018 (5), p. 31ff



(Bright-field, stained specimen: courtesy of Prof. Dr. Rüdiger Waldherr)

Fig. 11.54 Tumor cells

11.8 Casts: Overview

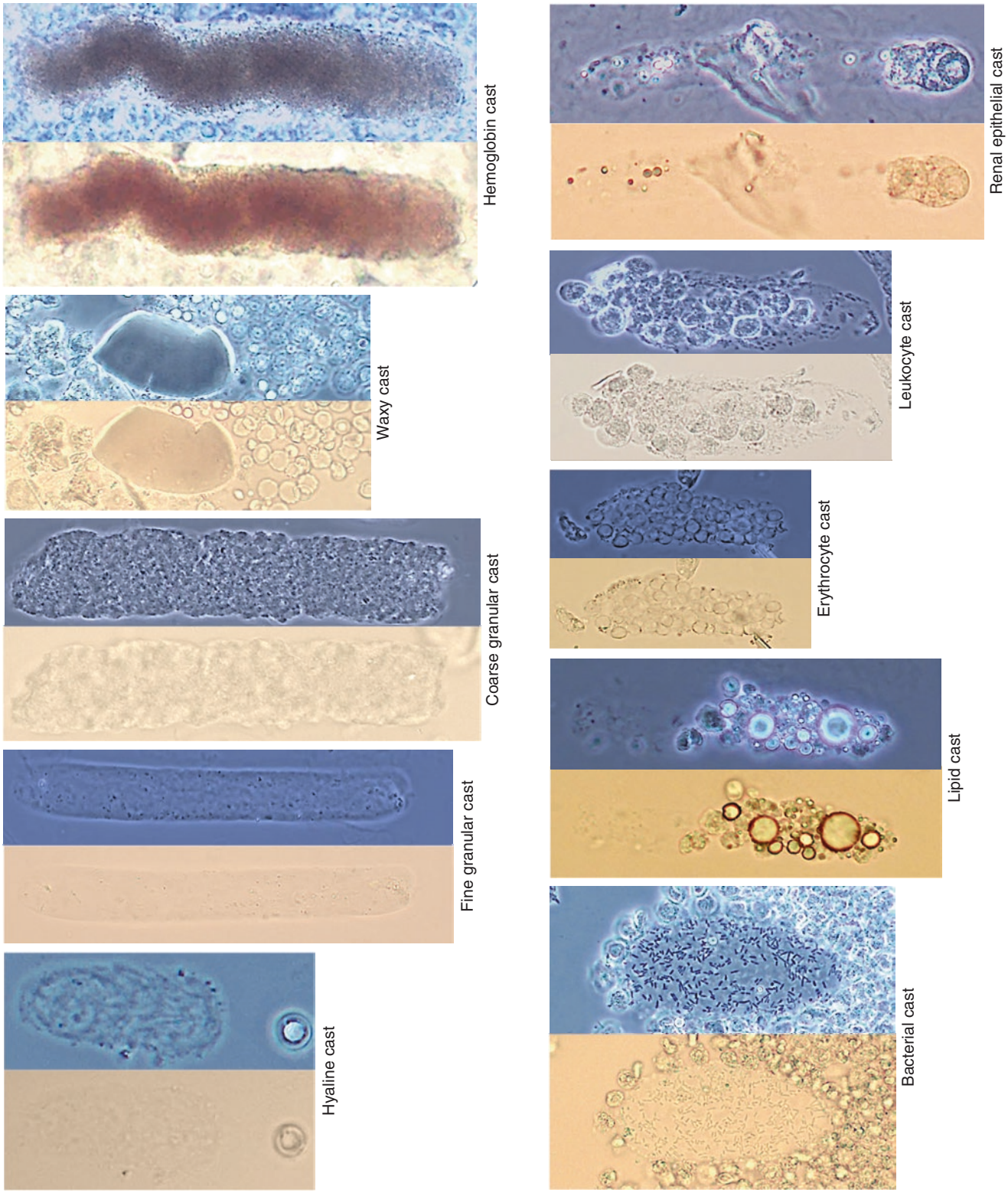


Fig. 11.55 Overview of urinary casts: *left*, bright-field and *right*, phase-contrast image

11.8.1 Pseudocasts = Mucus Threads

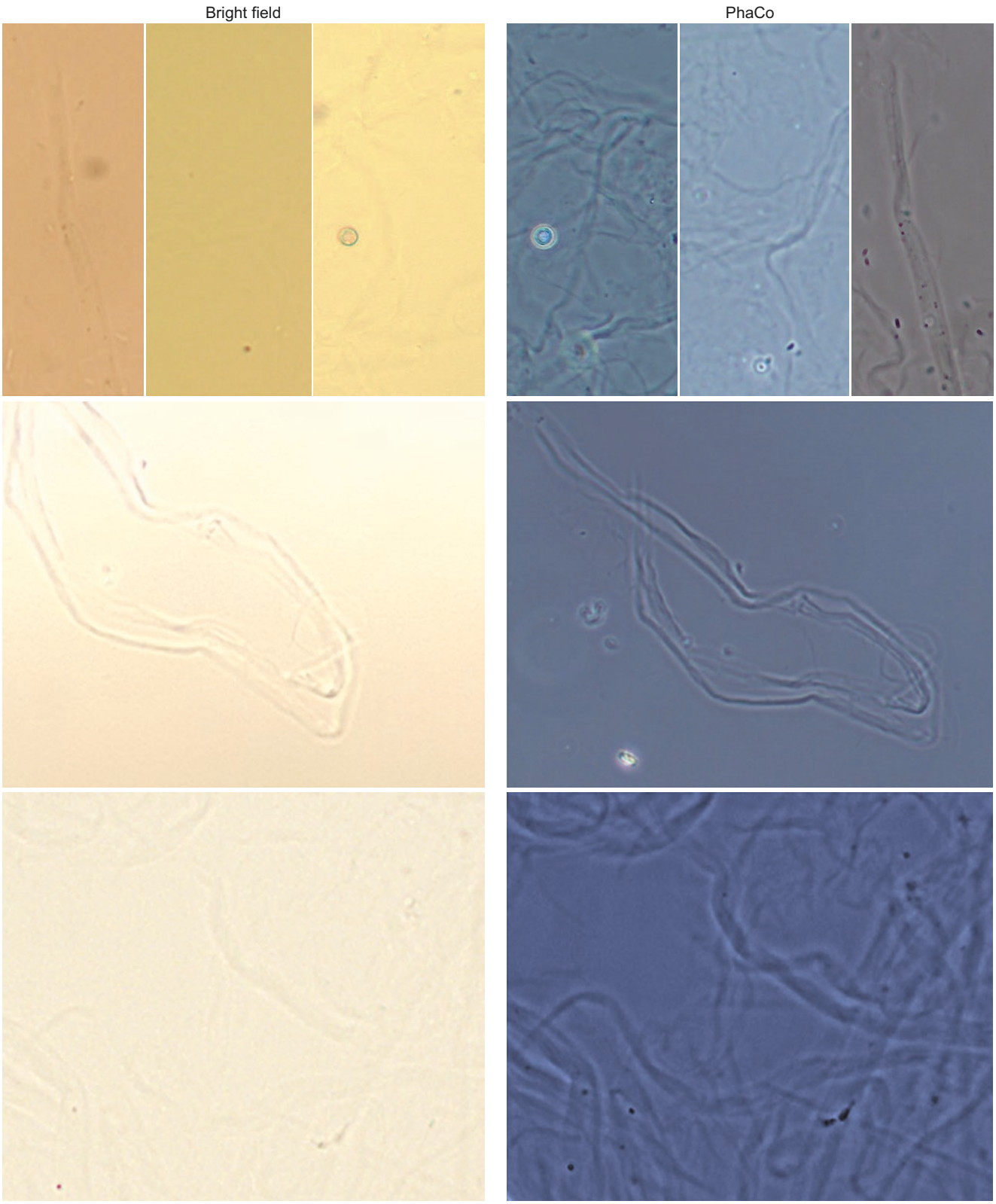


Fig. 11.56 Like hyaline casts, mucus threads are not, or only poorly, visible in bright-field mode

11.8.2 Hyaline Casts

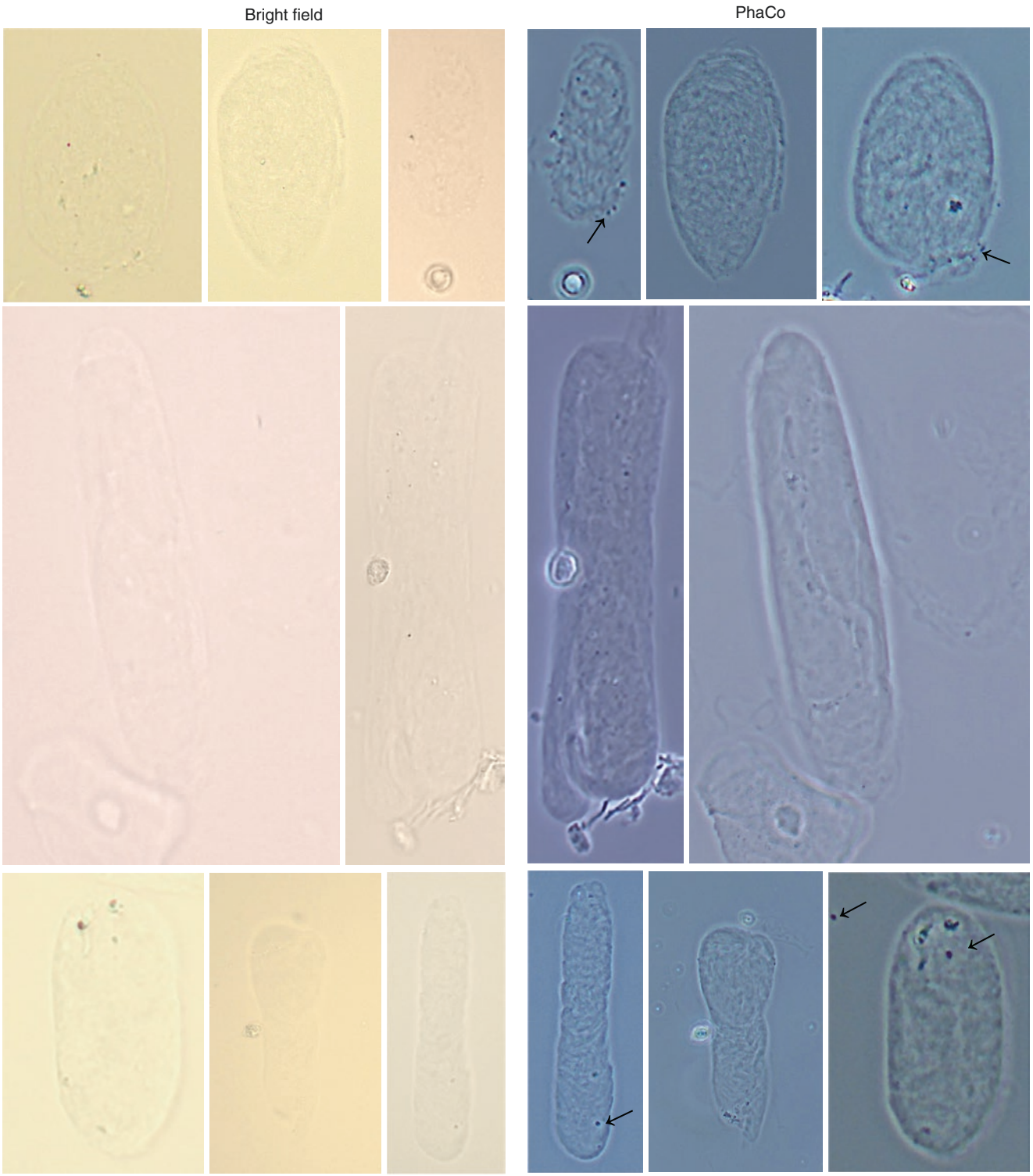


Fig. 11.57 Cast volume varies considerably. Bacteria (→) and artifacts lie partially on the hyaline matrix. These deposits of constituents need to be distinguished from granular casts and bacterial casts. The differentiation of a hyaline cast from a waxy cast is diagnostically important, since a waxy cast should be regarded as pathological, in contrast to a hyaline cast. Differentiation of the two cast types is straightforward if one takes into account that hyaline casts, in contrast to waxy casts, are virtually undetectable in bright-field microscopy

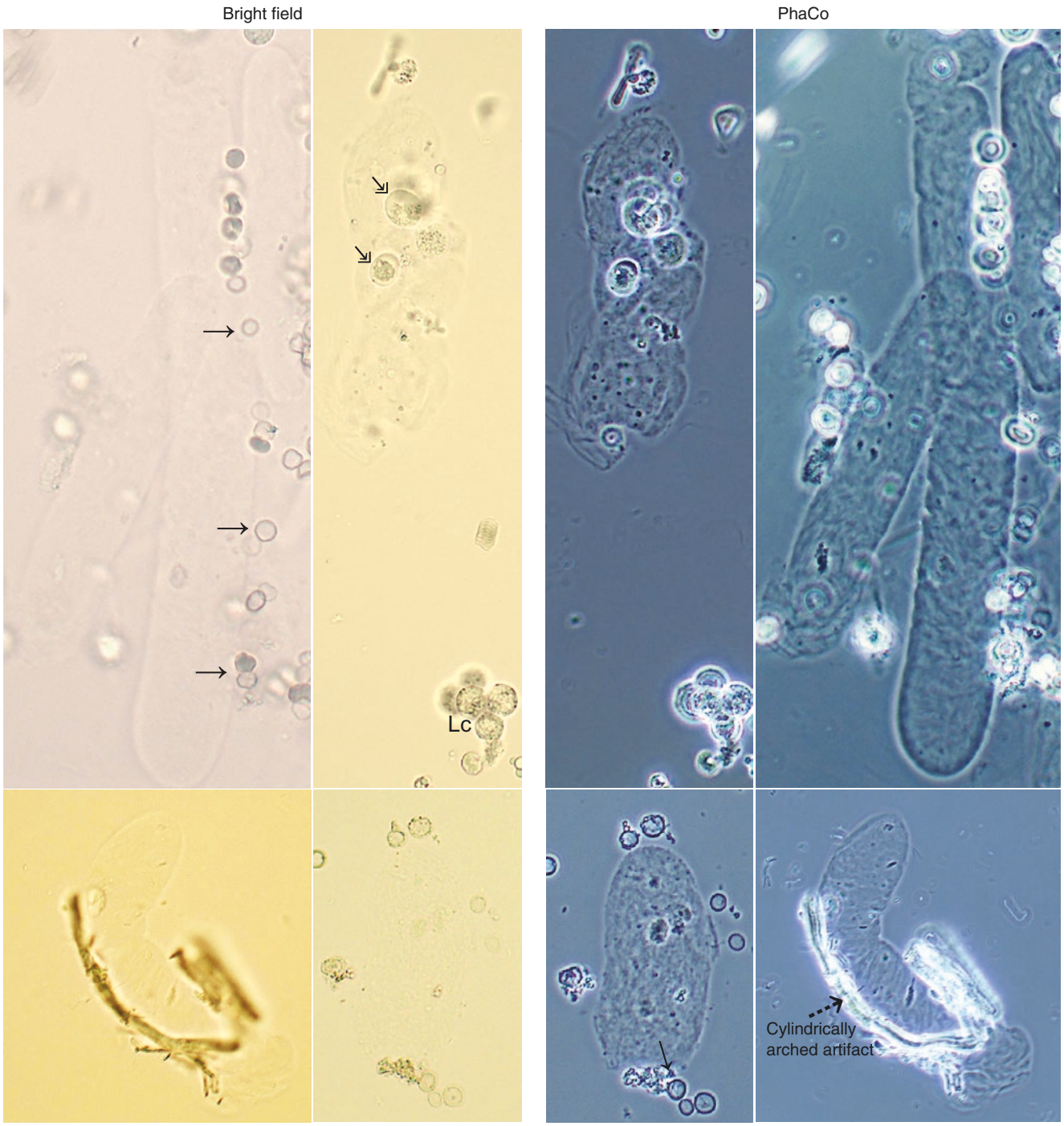
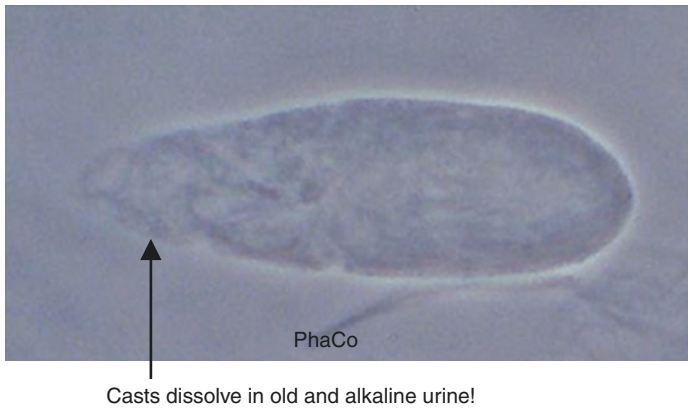


Fig. 11.58 Erythrocyte deposits (→) on the hyaline casts must not be confused with erythrocyte casts. Leukocyte deposits (↘) must not be confused with leukocyte casts

11.8.3 Old Casts



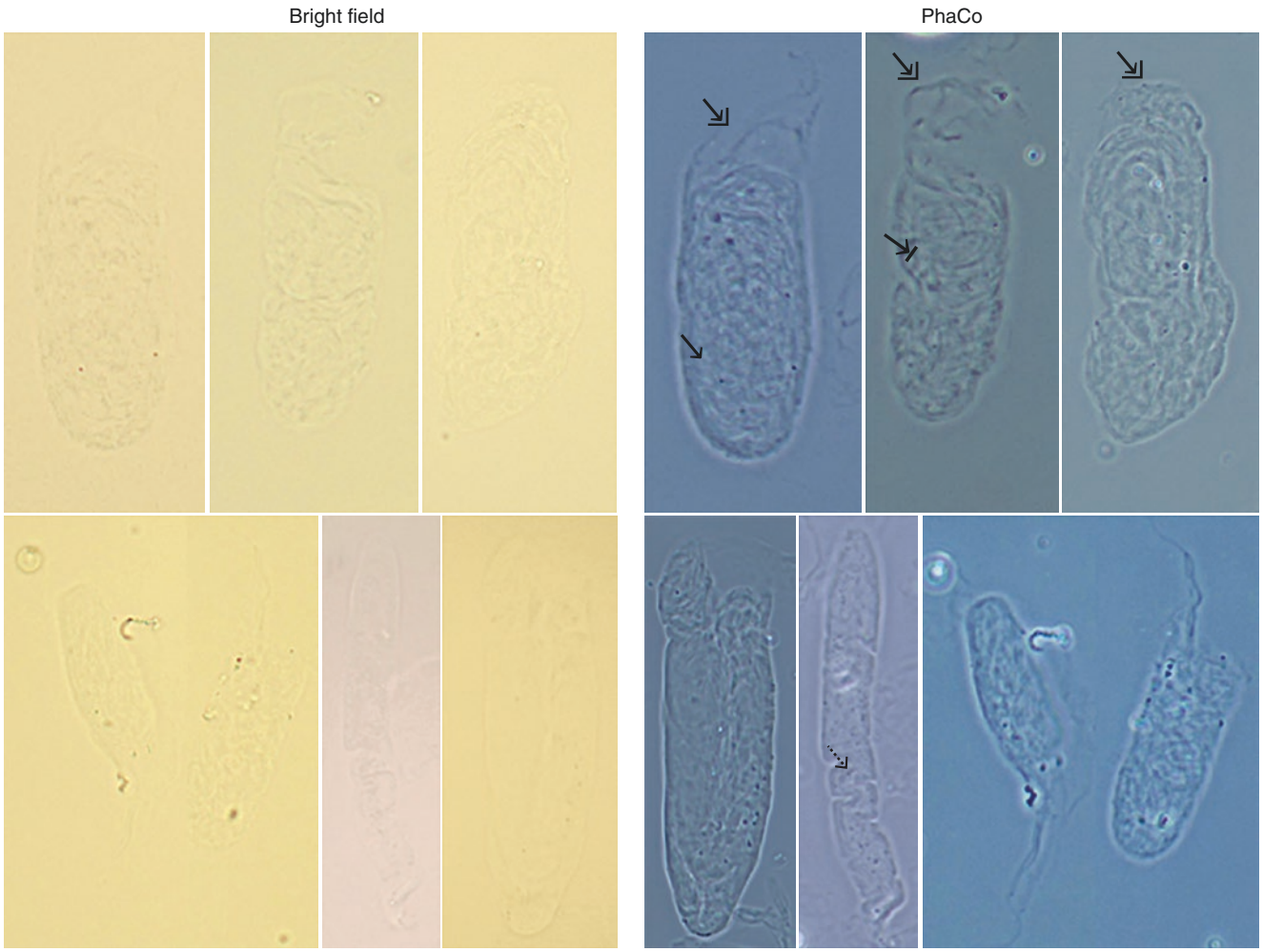


Fig. 11.59 Typical signs of "aging" include frayed processes (⇲) at the ends of the casts. The hyaline substance can have a fine (↘) or coarse (⇥) structure. The hyaline matrix also dissolves at the center of the cast (⇢) as a sign of degeneration. Hyaline casts are virtually invisible in bright-field mode. Tip for bright-field microscopy: close the aperture diaphragm on the condenser somewhat to contrast the image!

11.8.4 Waxy Casts

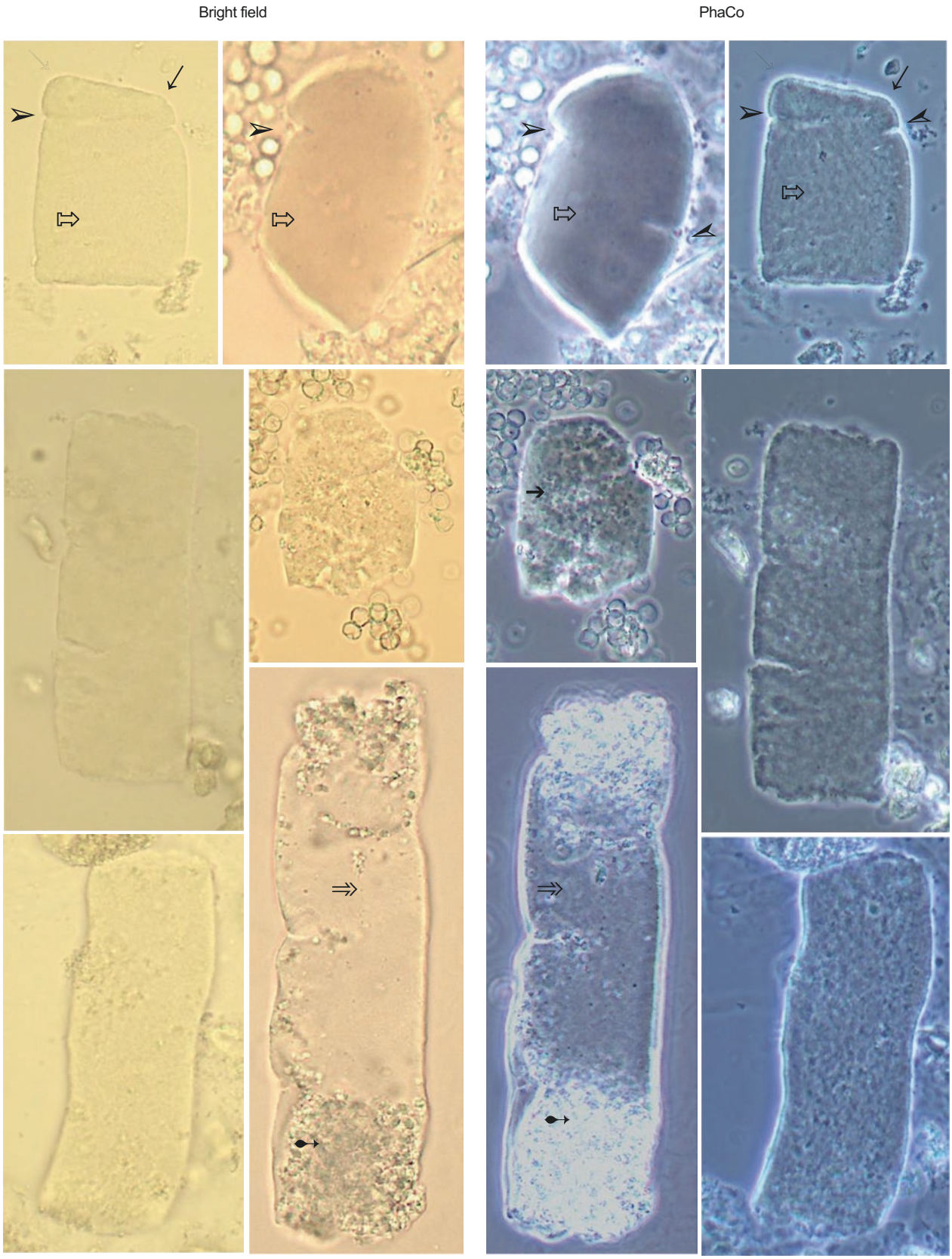


Fig. 11.60 The appearance of waxy casts varies: angular ends (→) and lateral indentations (➢) are typical. The interior can appear milky/cloudy (⇨) or slightly marbled (➔). There are also mixed forms, i.e., the cast is partly waxy (⇛) and partly granular (➻)

11.8.5 Granular Casts

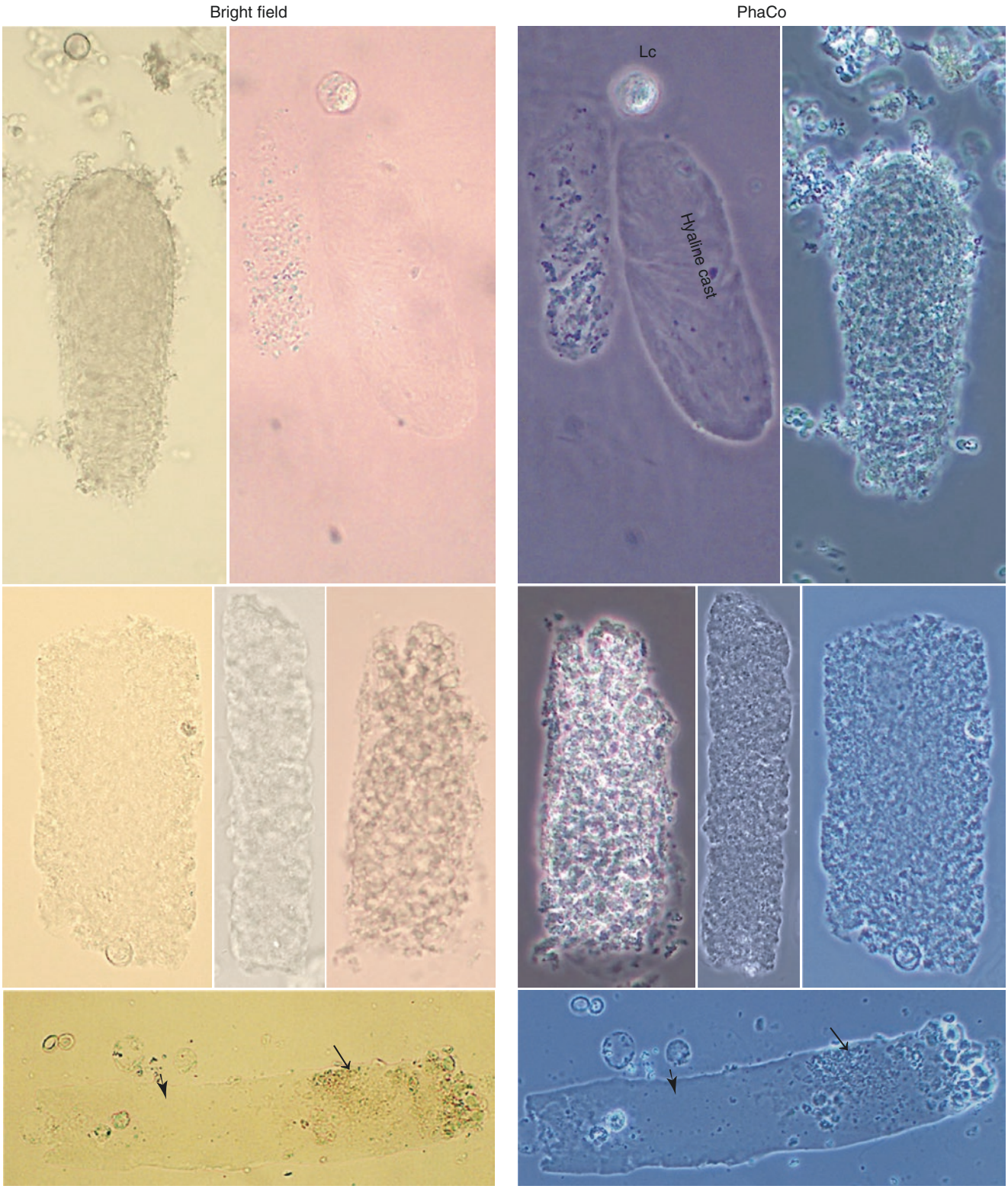


Fig. 11.61 Granular casts have granulation that fills the cast either sparsely or densely and compactly. Mixed casts: casts may also be only partially granular, as shown in the example below: half granular (→) and half waxy (➛)

11.8.6 Erythrocyte Casts

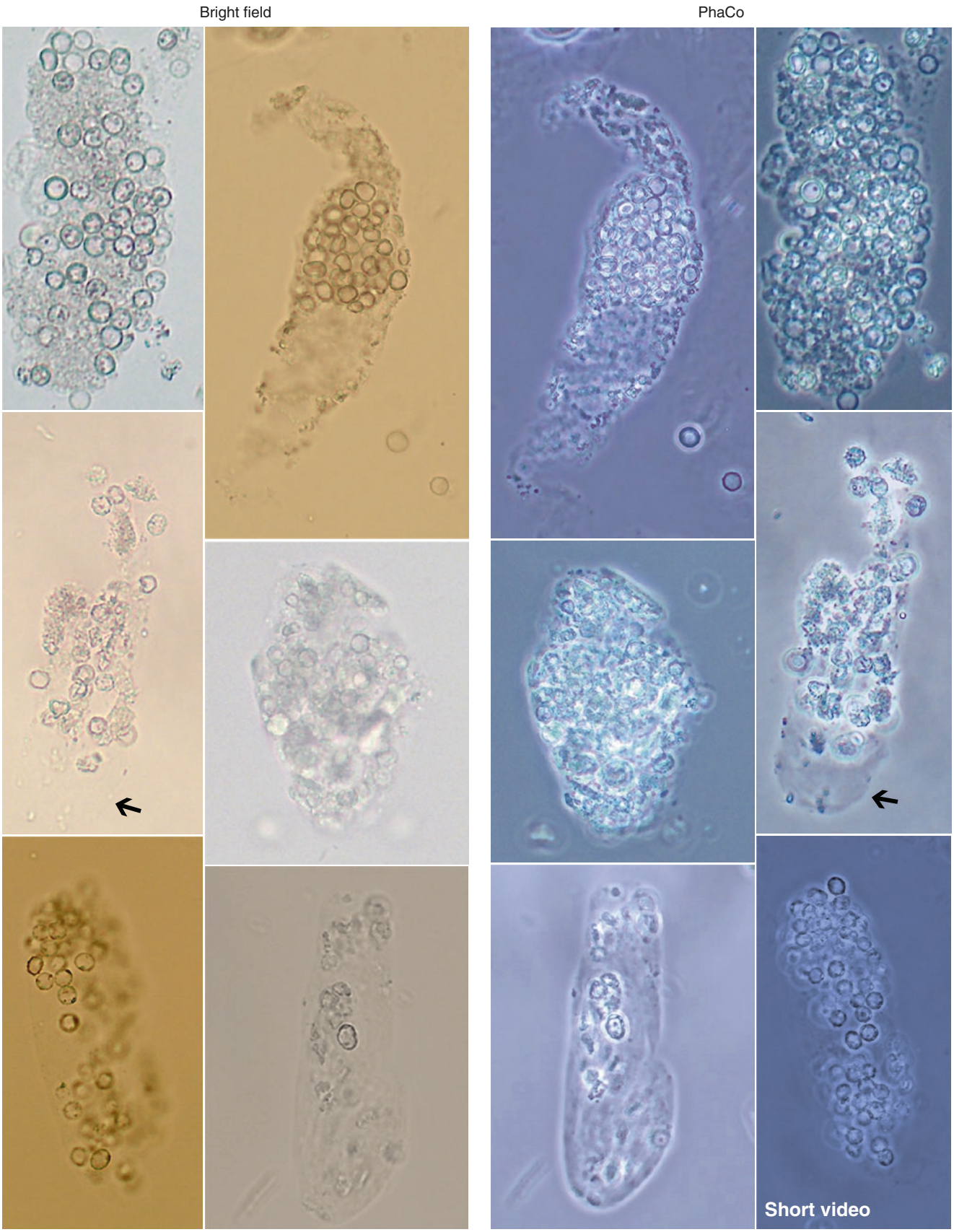


Fig. 11.62 The density to which erythrocytes are packed and the volume of erythrocyte casts vary considerably. The inclusion of erythrocytes in a cast can only be clearly visualized using the phase-contrast technique (comparison ➔). (see Video 11.13)

11.8.7 Hemoglobin Casts

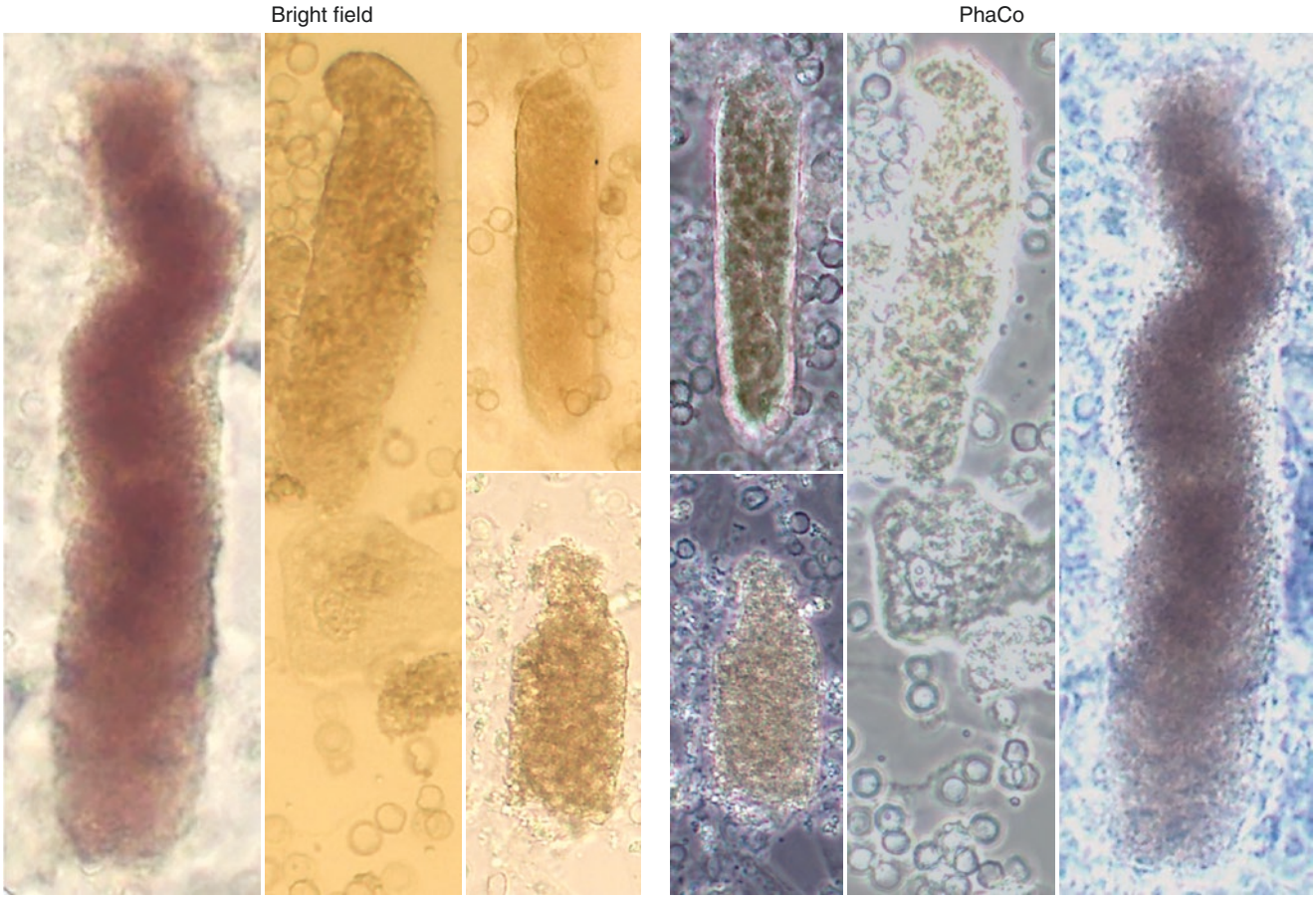


Fig. 11.63 Typical features include their inherent brown-red color and the granular—but not crystalline—filling of the cast

11.8.8 Leukocyte Casts

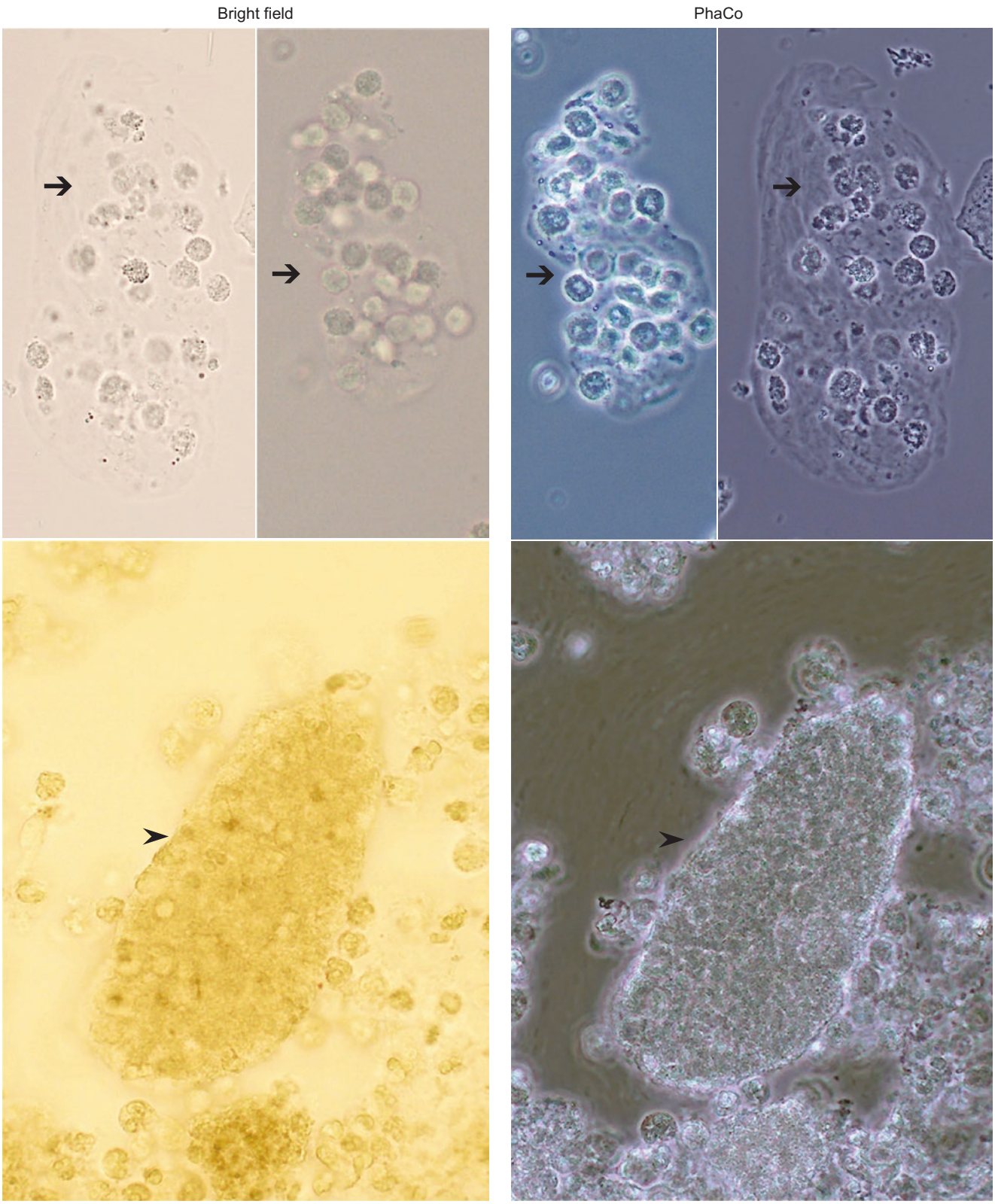


Fig. 11.64 The density of packing varies greatly, ranging from individually embedded leukocytes (➔) to densely packed leukocytes (➤)

11.8.9 Renal Epithelial Casts

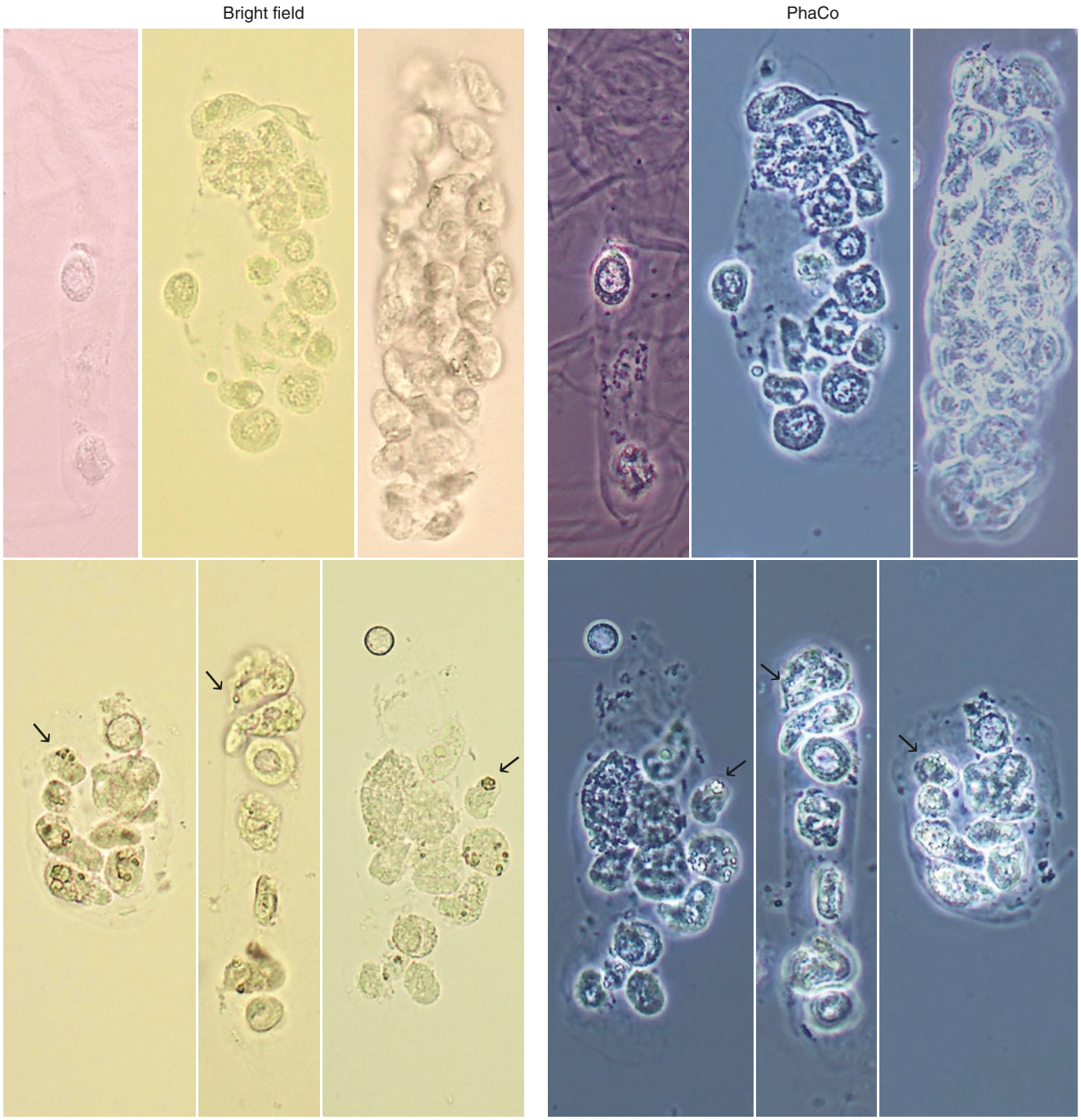


Fig. 11.65 The number of epithelial cells in a cast varies greatly—from a single epithelial cell to many densely packed epithelial cells. Epithelial casts are difficult to distinguish from leukocyte casts; they are easier to differentiate when renal epithelial cells contain fat droplets (↘)

11.8.10 Mixed Cell Casts

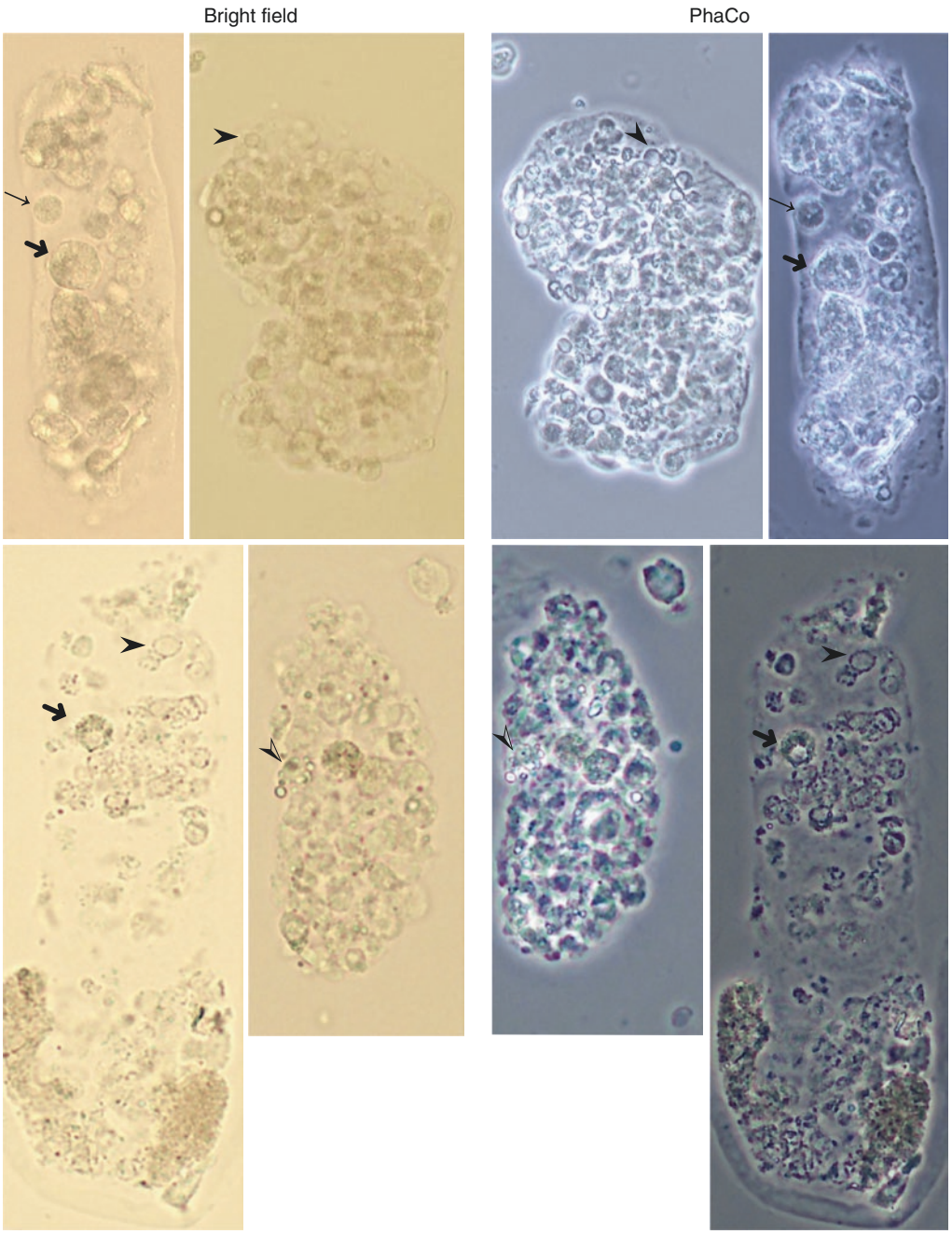


Fig. 11.66 In addition to larger round cells (renal epithelial cells ➔), smaller cells (leukocytes → and erythrocytes ➤) can also be seen in the cast. The oval fat bodies (➢) lie next to cells that, unfortunately, cannot be clearly differentiated here

11.8.11 Microscopy Technique: E.g., Casts

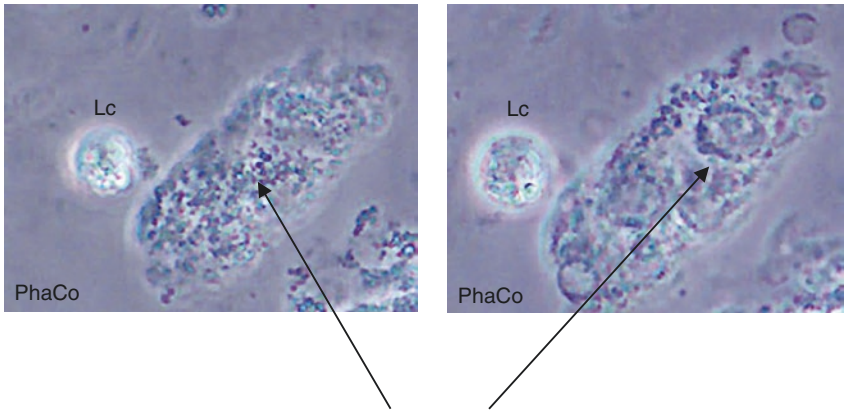


Fig. 11.67 Tip: when performing microscopy analysis, the micrometer knob should be constantly turned! This will reveal that the granular cast is a mixed cell cast

11.8.12 Oval Fat Body Casts

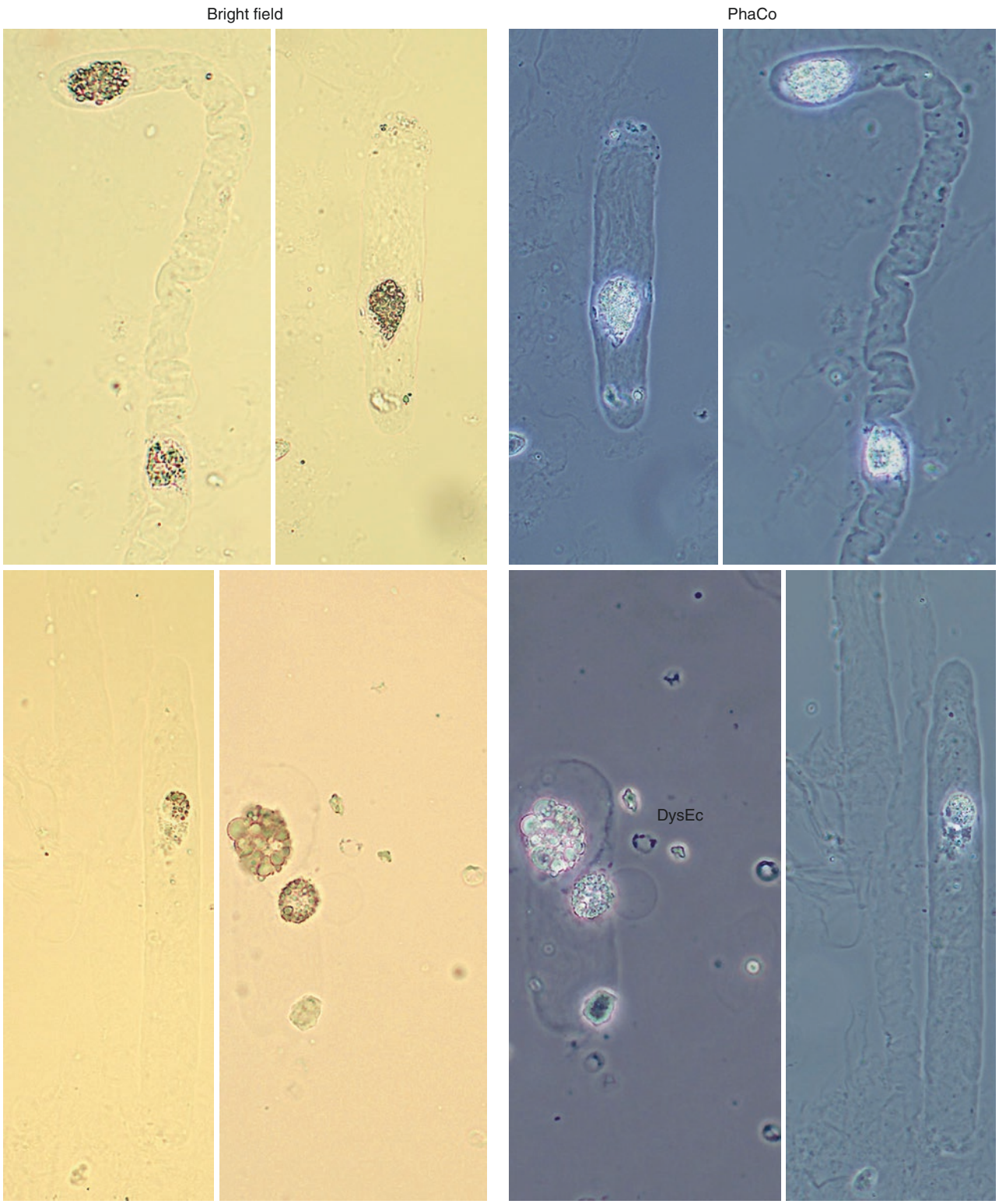


Fig. 11.68 Oval fat body casts

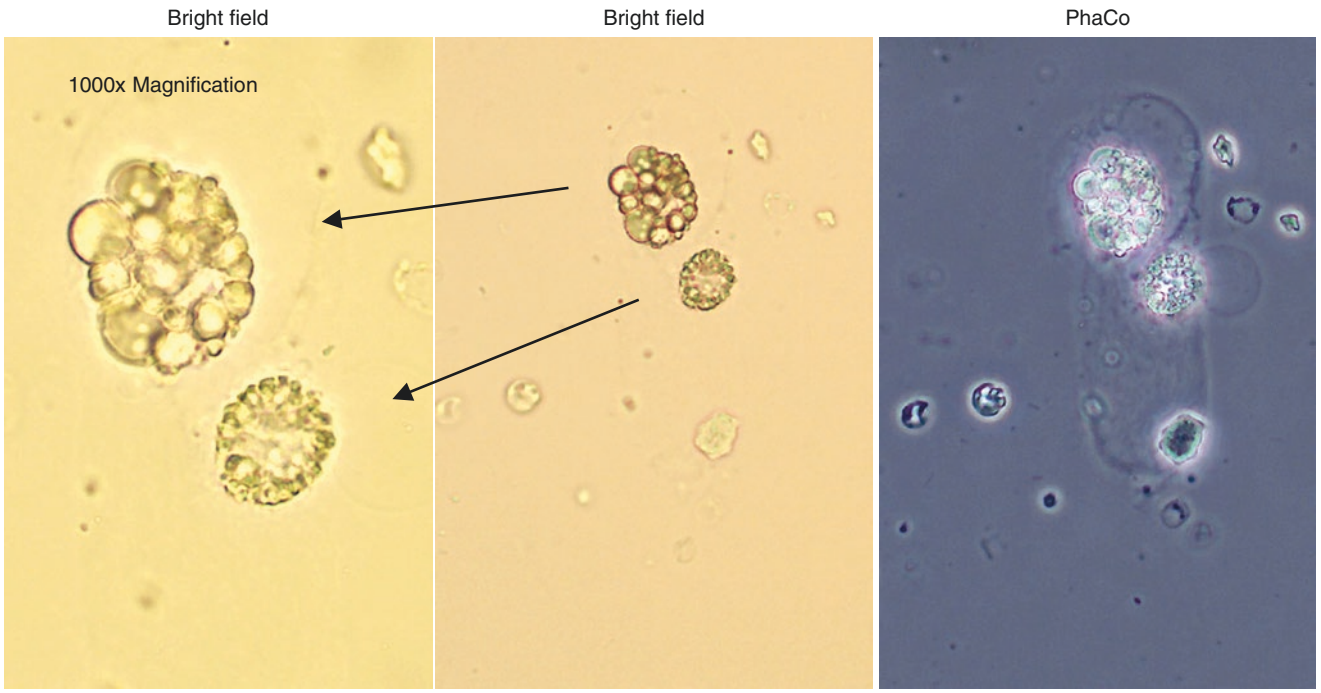


Fig. 11.69 Oval fat body casts at 1000× and 400× magnification

11.8.13 Lipid Casts

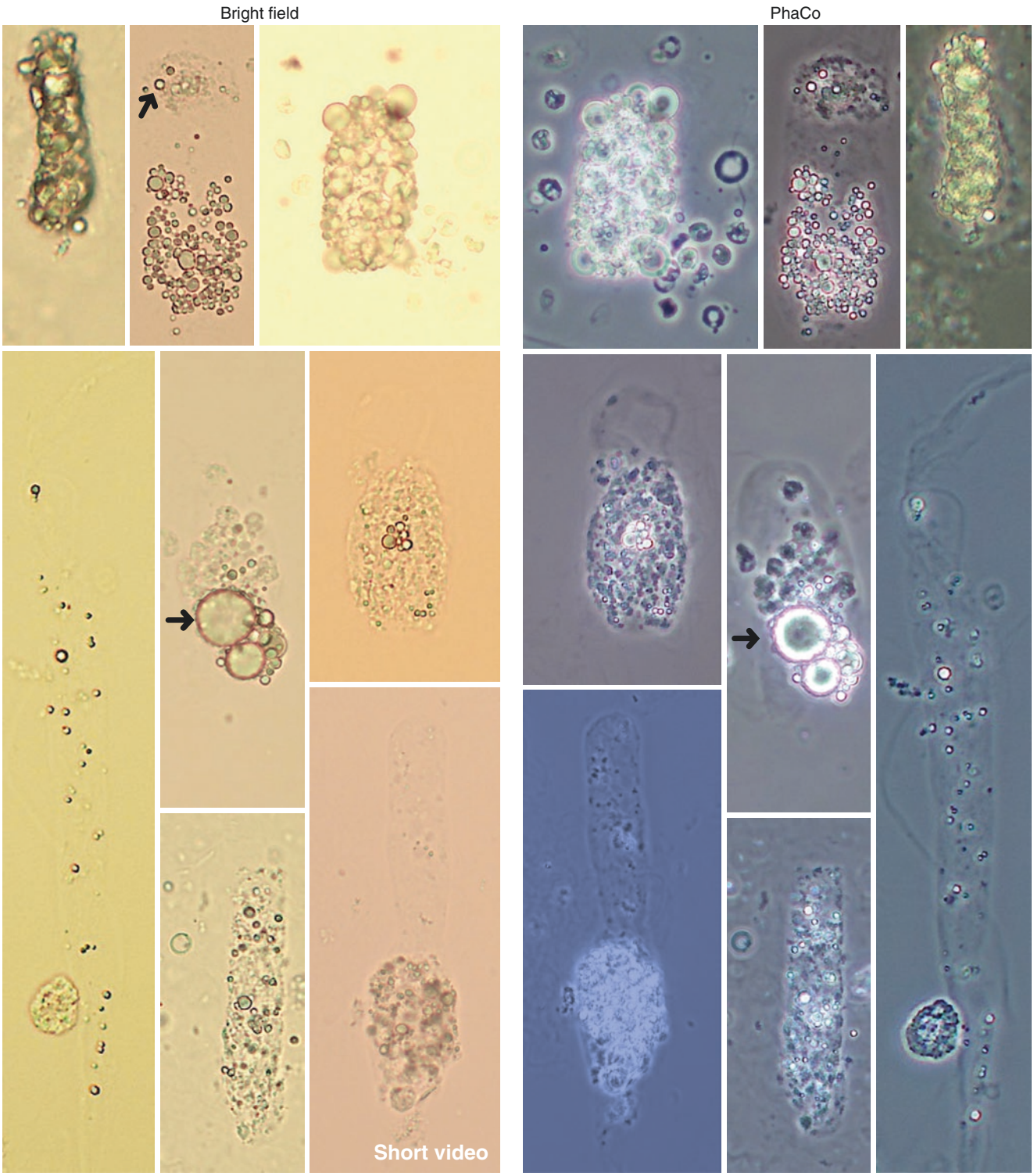


Fig. 11.70 Lipid cast I: typical round, shiny fat particles of varying size and density are characteristic of the lipid cast. The difference in size (➔) of the fat inclusions is considerable. (see Video 11.14)

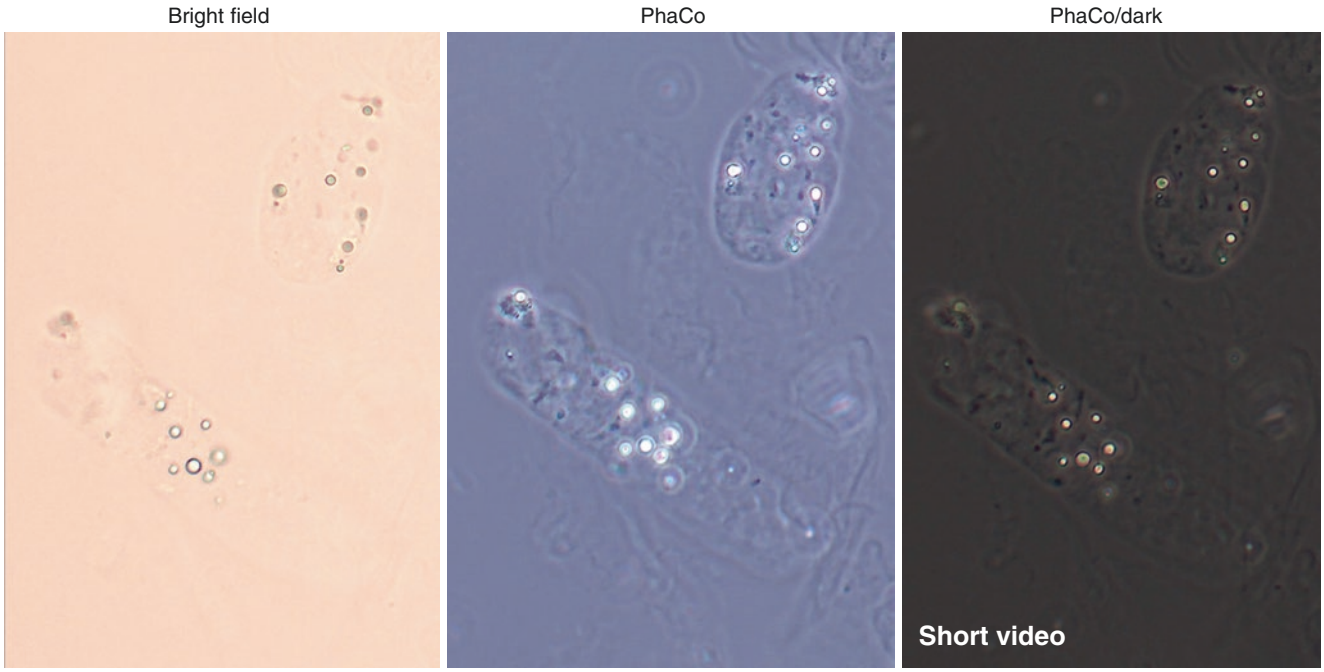


Fig. 11.71 Lipid cast II: the embedding of lipid particles in the hyaline matrix is readily visible in PhaCo. If there is uncertainty as to whether it is a fat particle, the bright yellow fat particles can be clearly differentiated by reducing the light (using the light dimmer on the microscope lamp). Conclusion: fat lights up clearly bright yellow. (see Video 11.15)

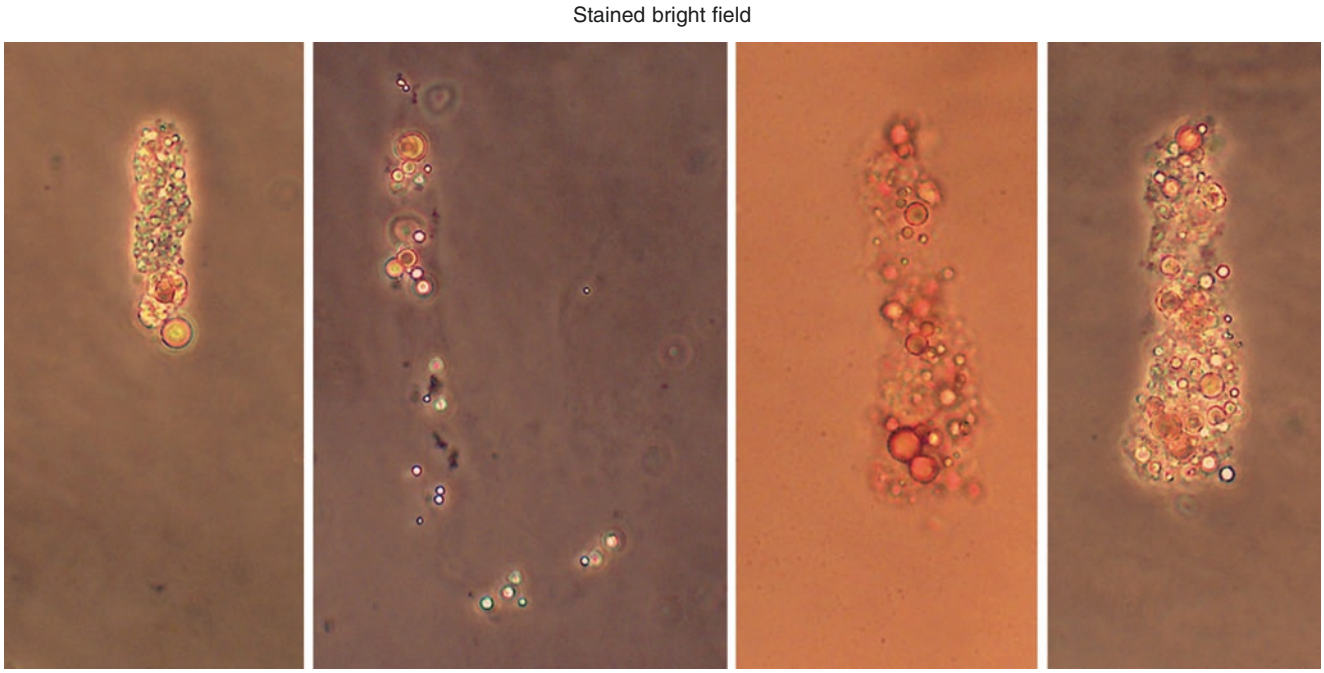


Fig. 11.72 Lipid casts, stained with Sudan IV

11.8.14 Bacterial Casts

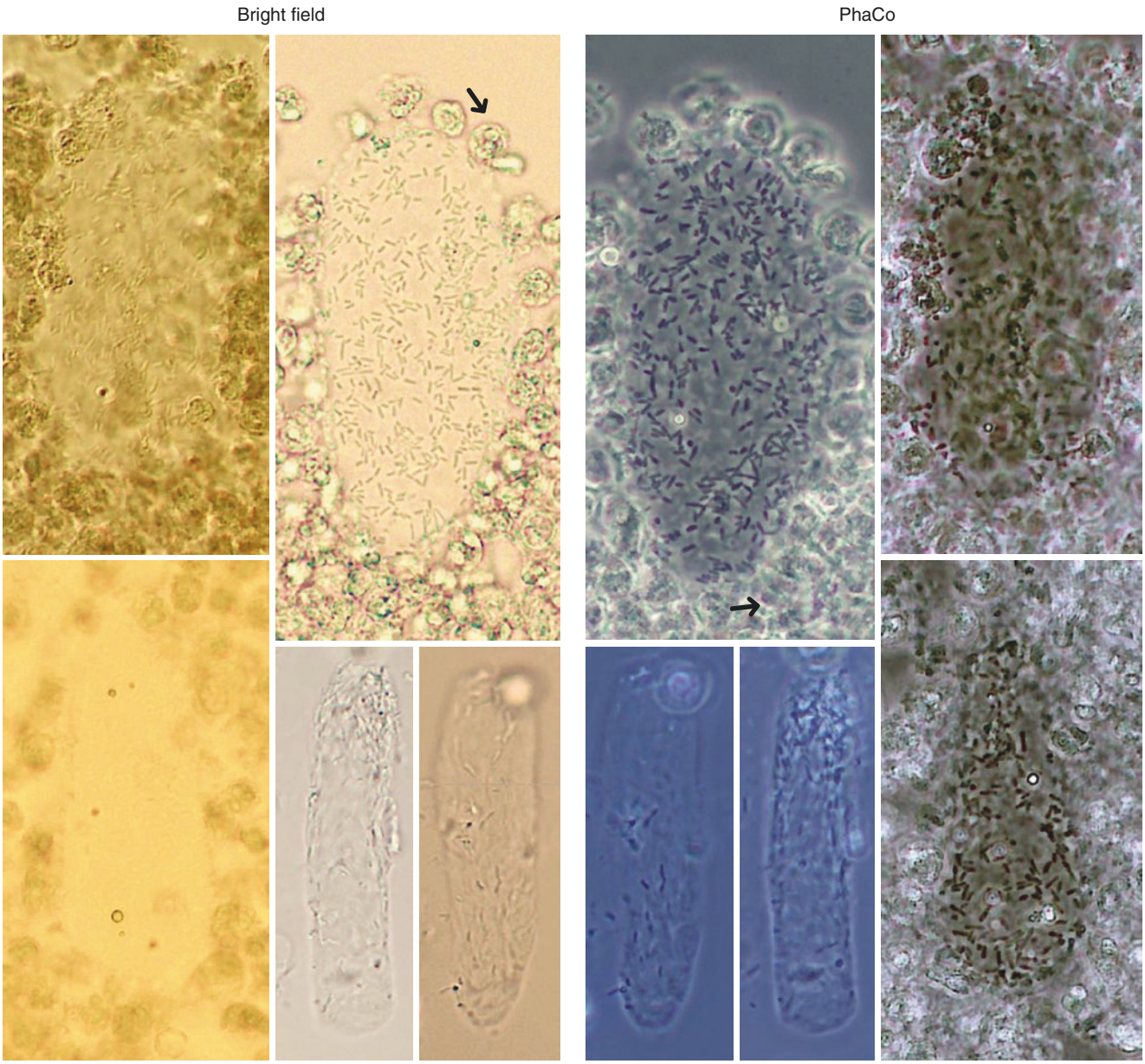


Fig. 11.73 Some of the bacterial casts lie embedded in numerous leukocytes (➔). Bacterial casts are much easier to identify in PhaCo than in bright-field mode

11.8.15 Long Casts: Erythrocyte Cast, Mixed Cell Cast and Oval Fat Body Cast

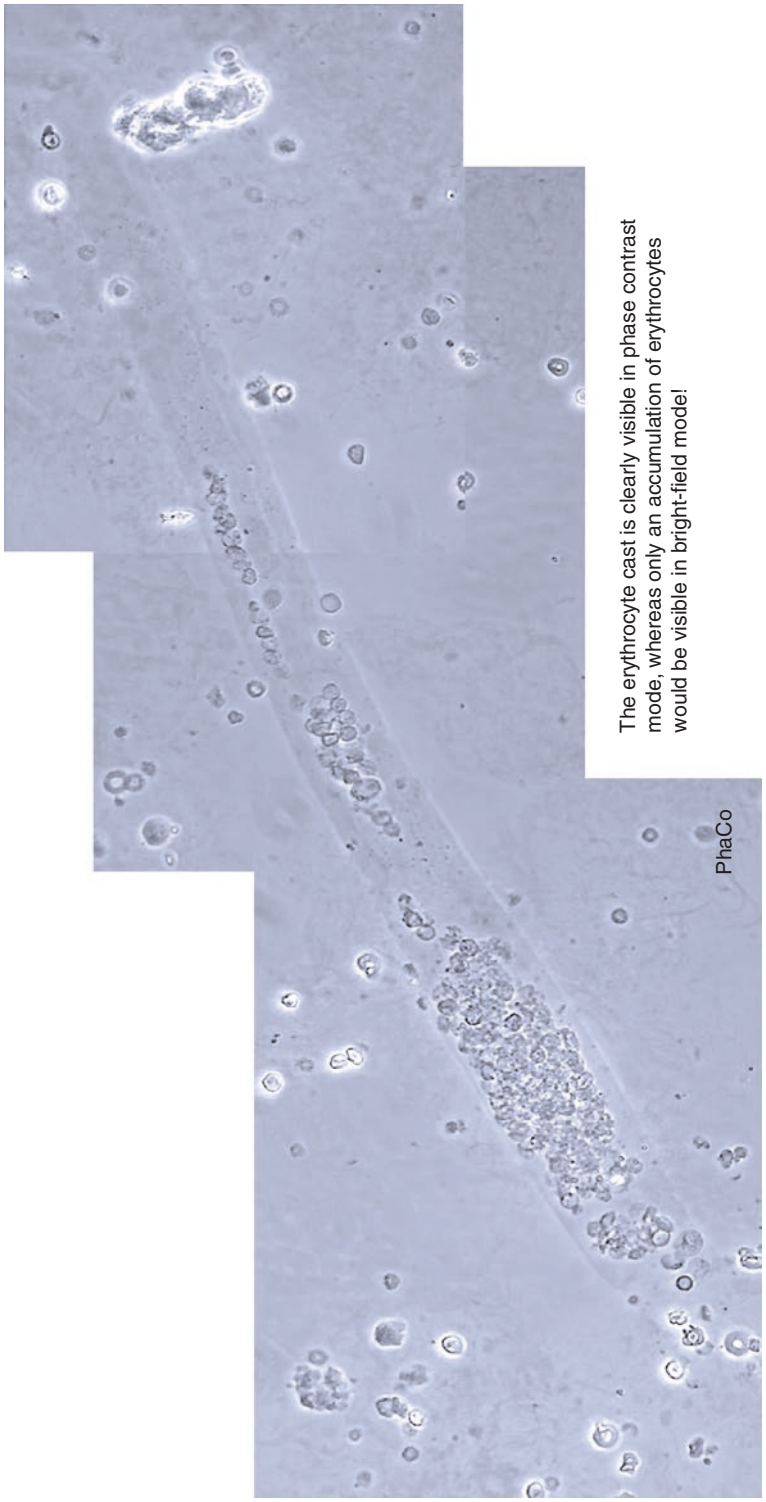


Fig. 11.74 Long casts: erythrocyte cast

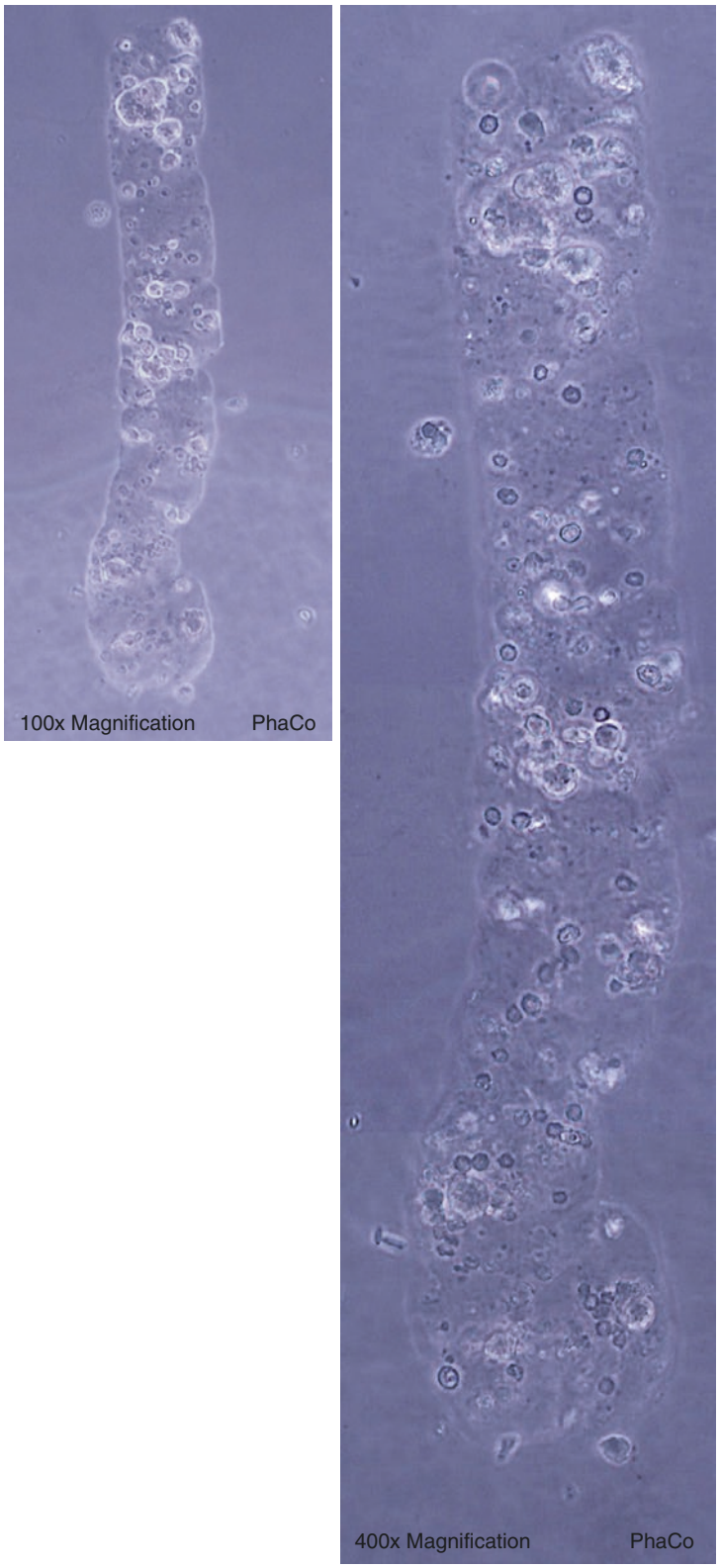


Fig. 11.75 Long casts: mixed cell cast (smaller cells such as erythrocytes and leukocytes, and larger cells such as renal epithelial cells)

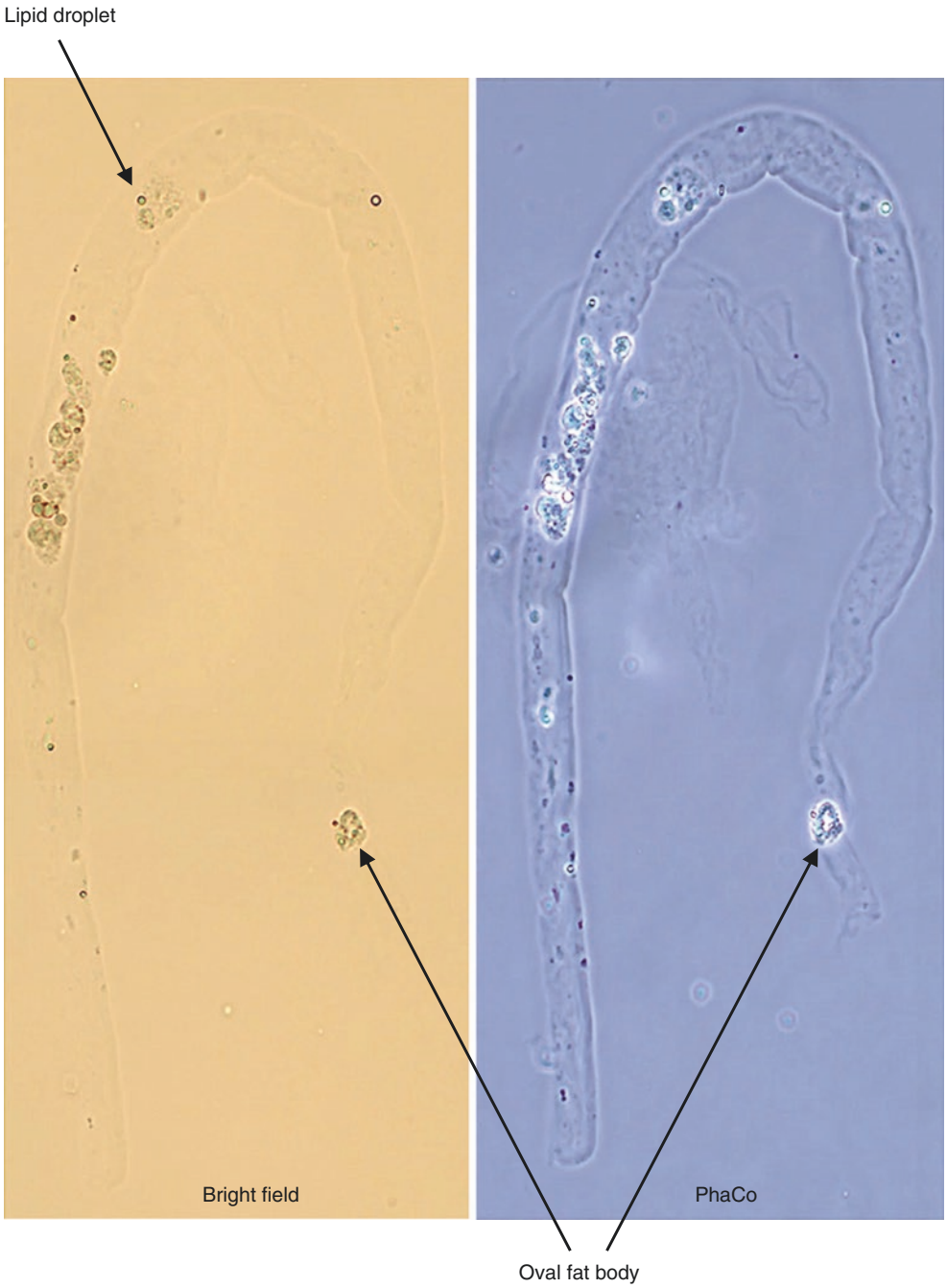


Fig. 11.76 Long casts: oval fat body cast

11.9 Bacteria

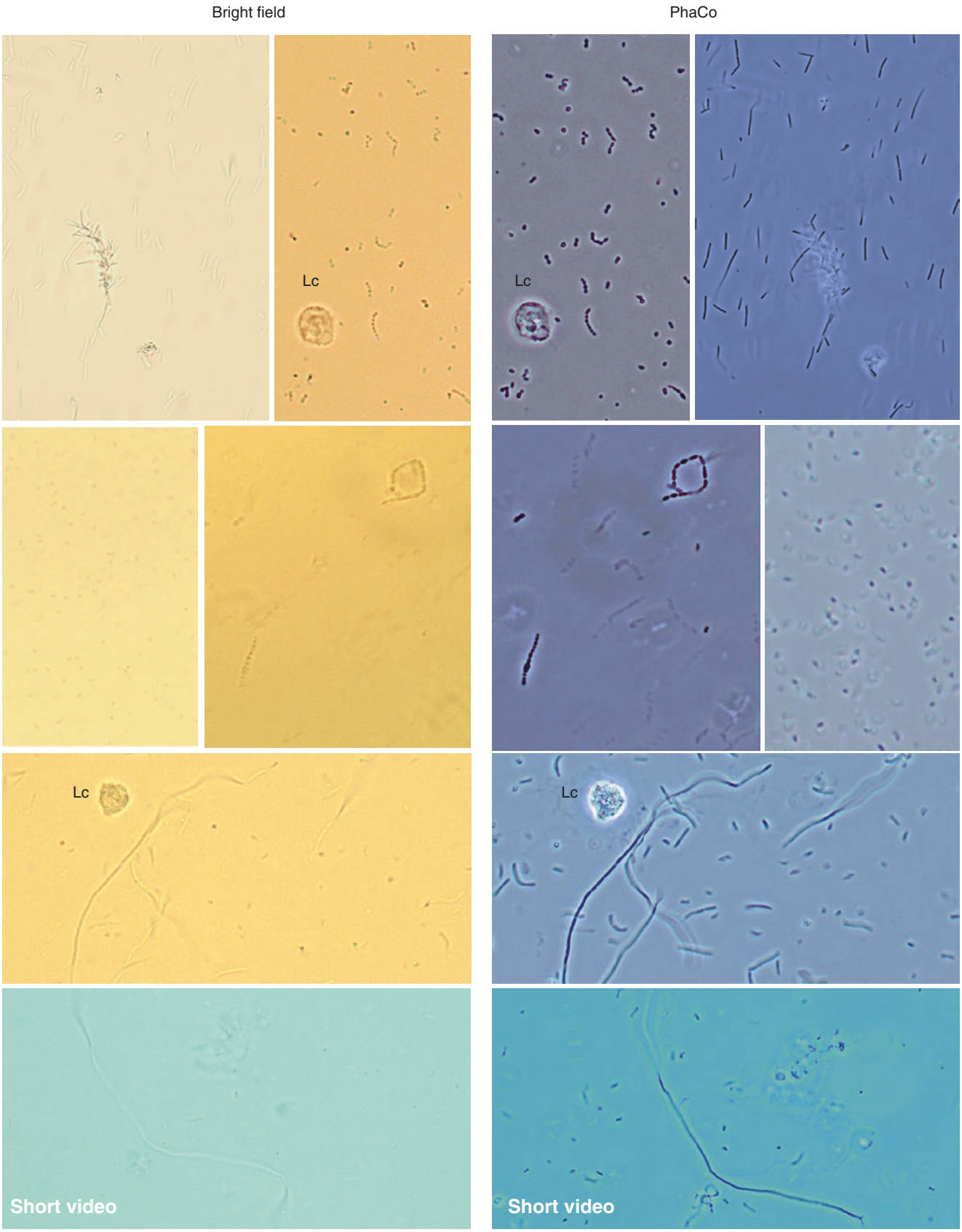


Fig. 11.77 Bacteria (rods, cocci) individually and in chains. Using the phase-contrast technique, the amount of bacteria in the unstained native specimen can be evaluated more reliably than in bright-field mode. (see Videos 11.16 and 11.17)

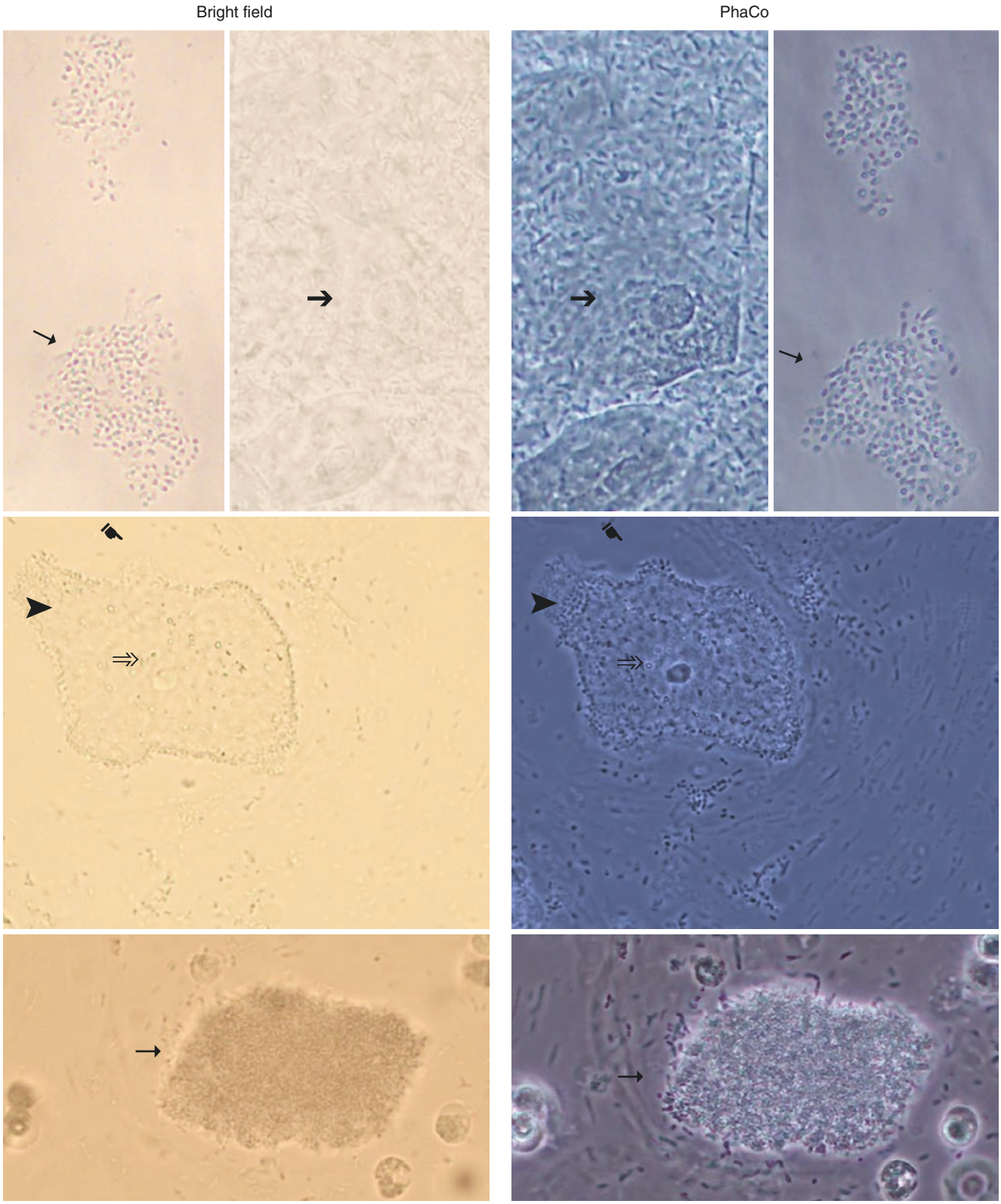


Fig. 11.78 Bacteria (rods, cocci) lying densely packed and in clusters (→). If bacterial density is high, other urine constituents such as epithelial cells (➔) appear blurred and undefined. The squamous epithelium (☛) has deposits of bacteria (➤), as well as a number of round and brightly luminescent fat droplets (⇛)

11.9.1 Semi-quantitative Bacterial Analysis

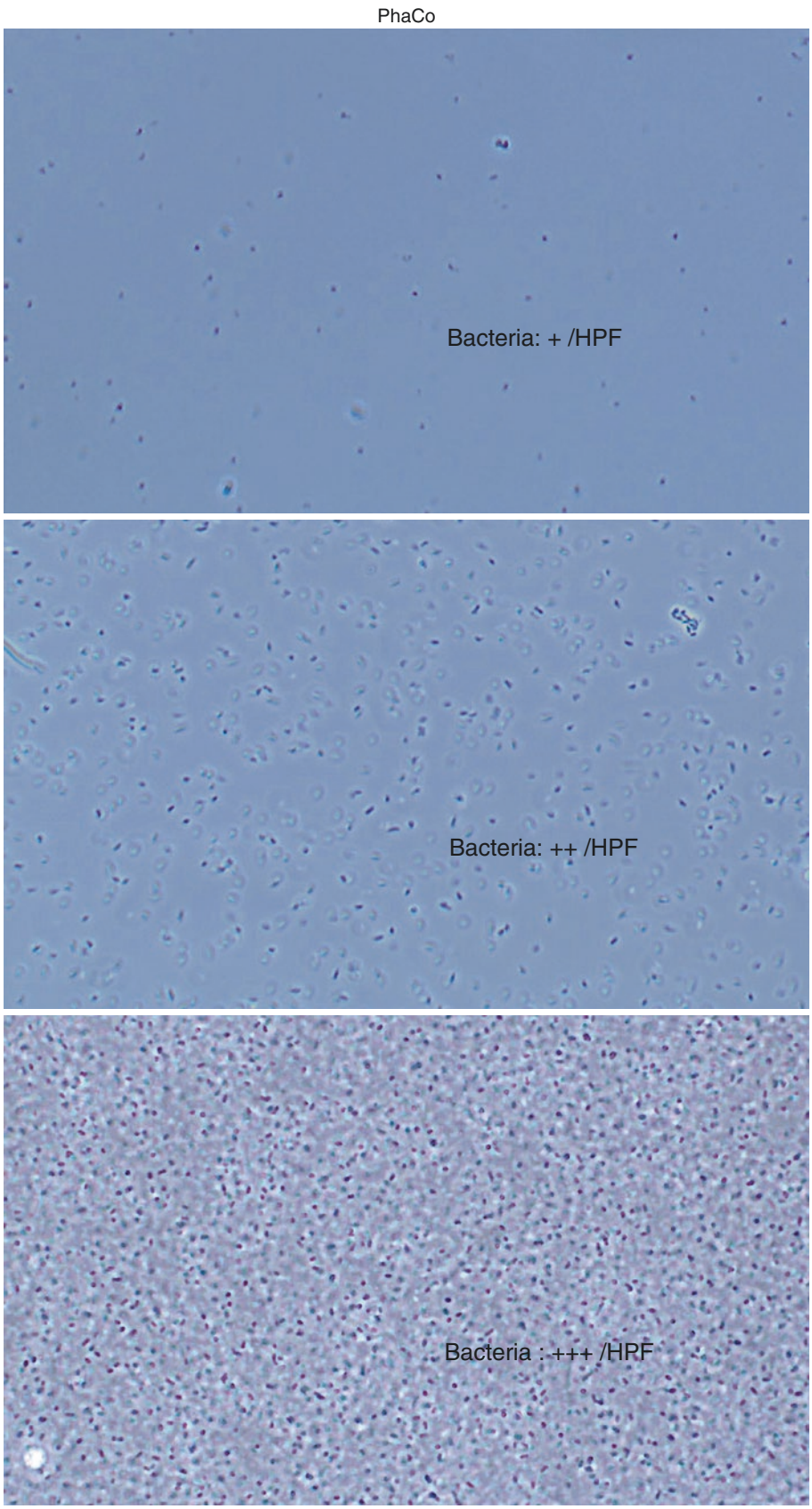


Fig. 11.79 Examples of semi-quantitative bacterial analysis. *Bacteria: +++/HPF* represents a uniform bacterial lawn

11.9.2 Discussion: Vaginal Swab

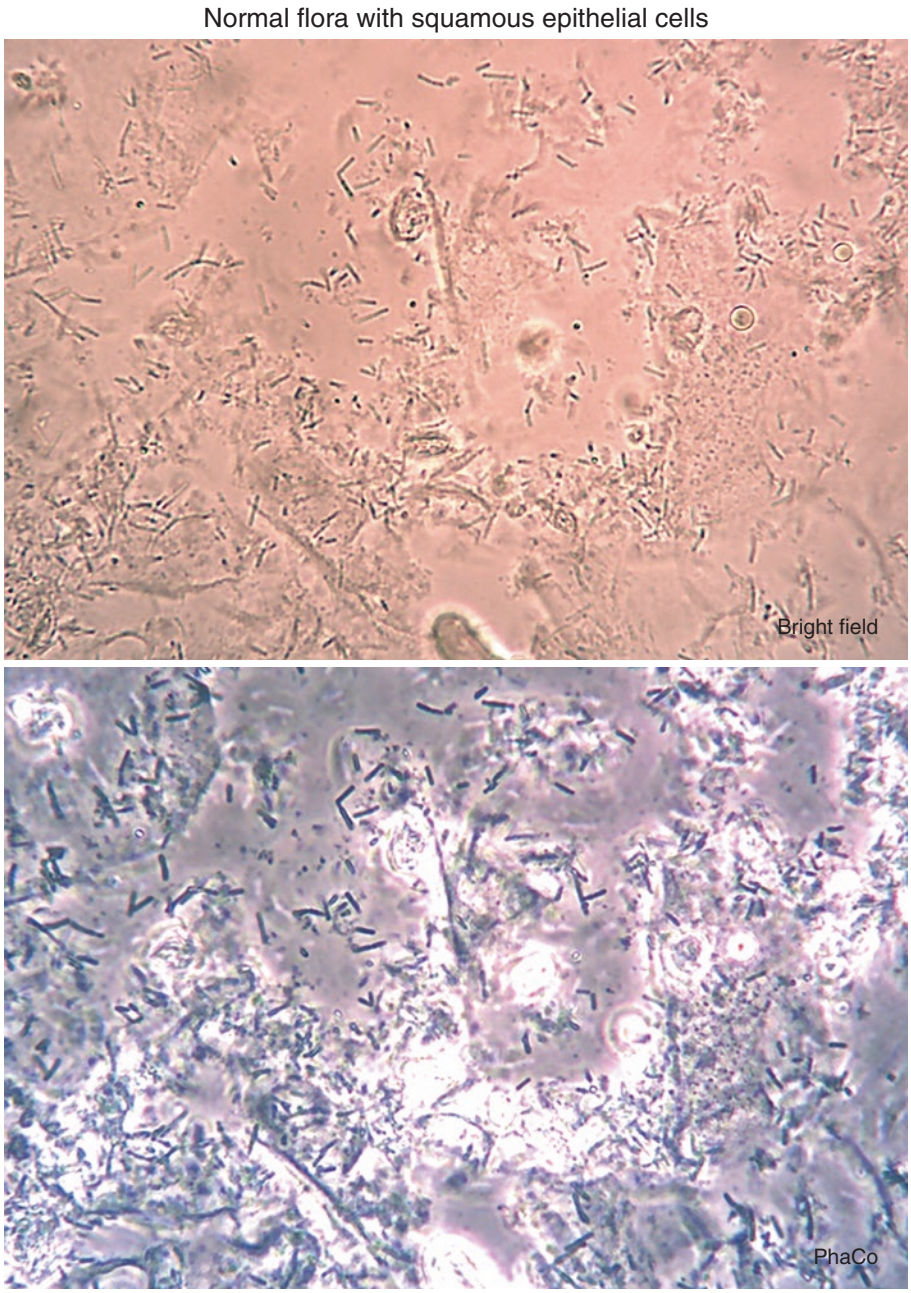


Fig. 11.80 Vaginal smear, normal flora with squamous epithelial cells, pH 4. The native specimen was mixed with 0.9 NaCl. Numerous squamous epithelial cells and Döderlein bacilli are visible in the vaginal swab. Quantities of vaginal secretion may also be found in urine

11.9.3 Discussion: Bacteriuria and Fecal Material

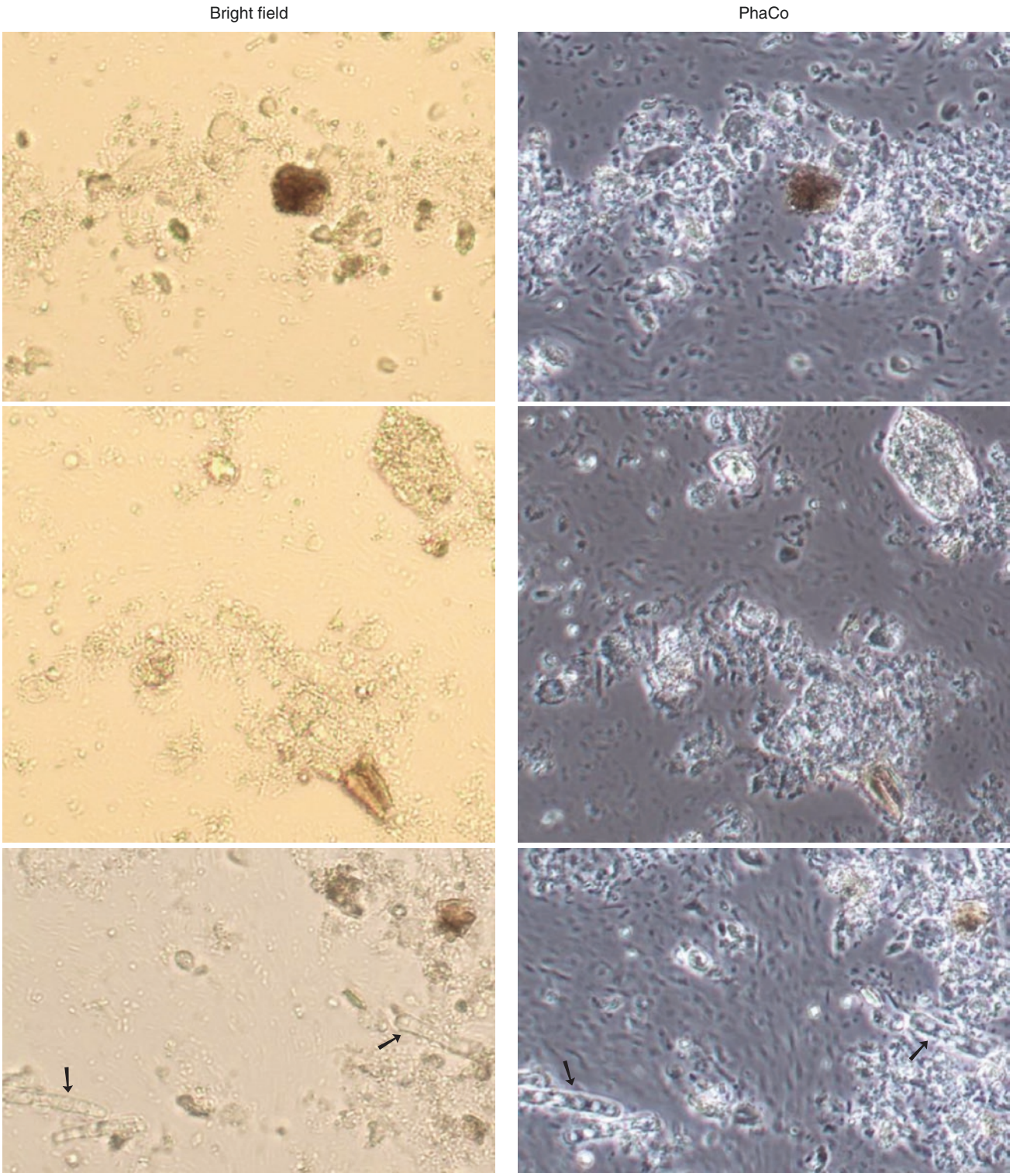


Fig. 11.81 Brown fecal material must not be confused with uric acid crystals. Increased presence of brown and colorless (➙) fecal material and massive concomitant bacteriuria due to rectal carcinoma with urinary bladder invasion or bladder-intestinal fistulas

11.10 Spermatozoa

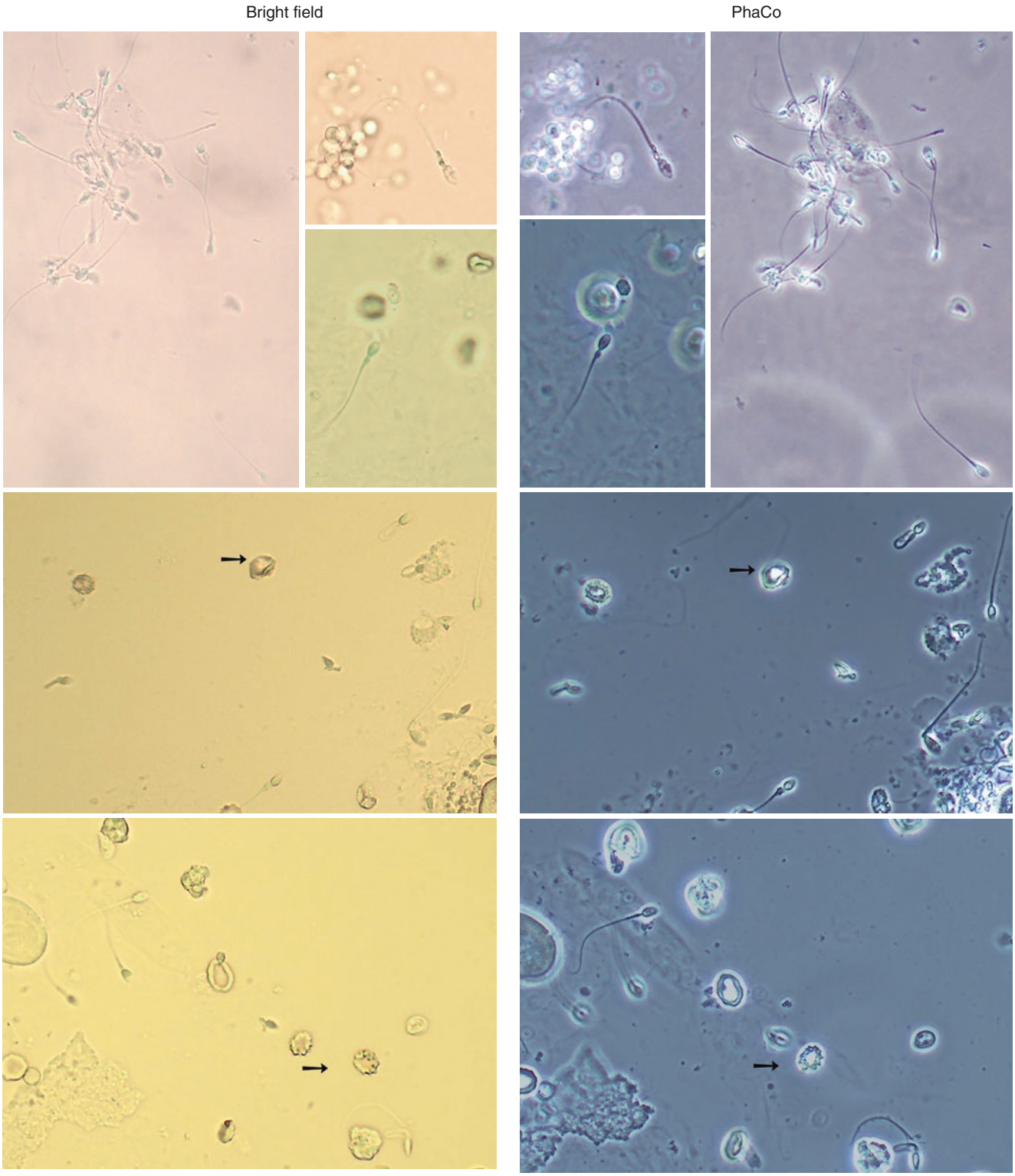


Fig. 11.82 Sperm are found individually or in clusters. The tail part of the sperm is easily overlooked in bright-field mode. The sperm heads are easily confused with yeast cells. Concomitant increased excretion of sperm and erythrocytes (➞) in the urine suggests hematospermia

11.11 Crystals: Overview

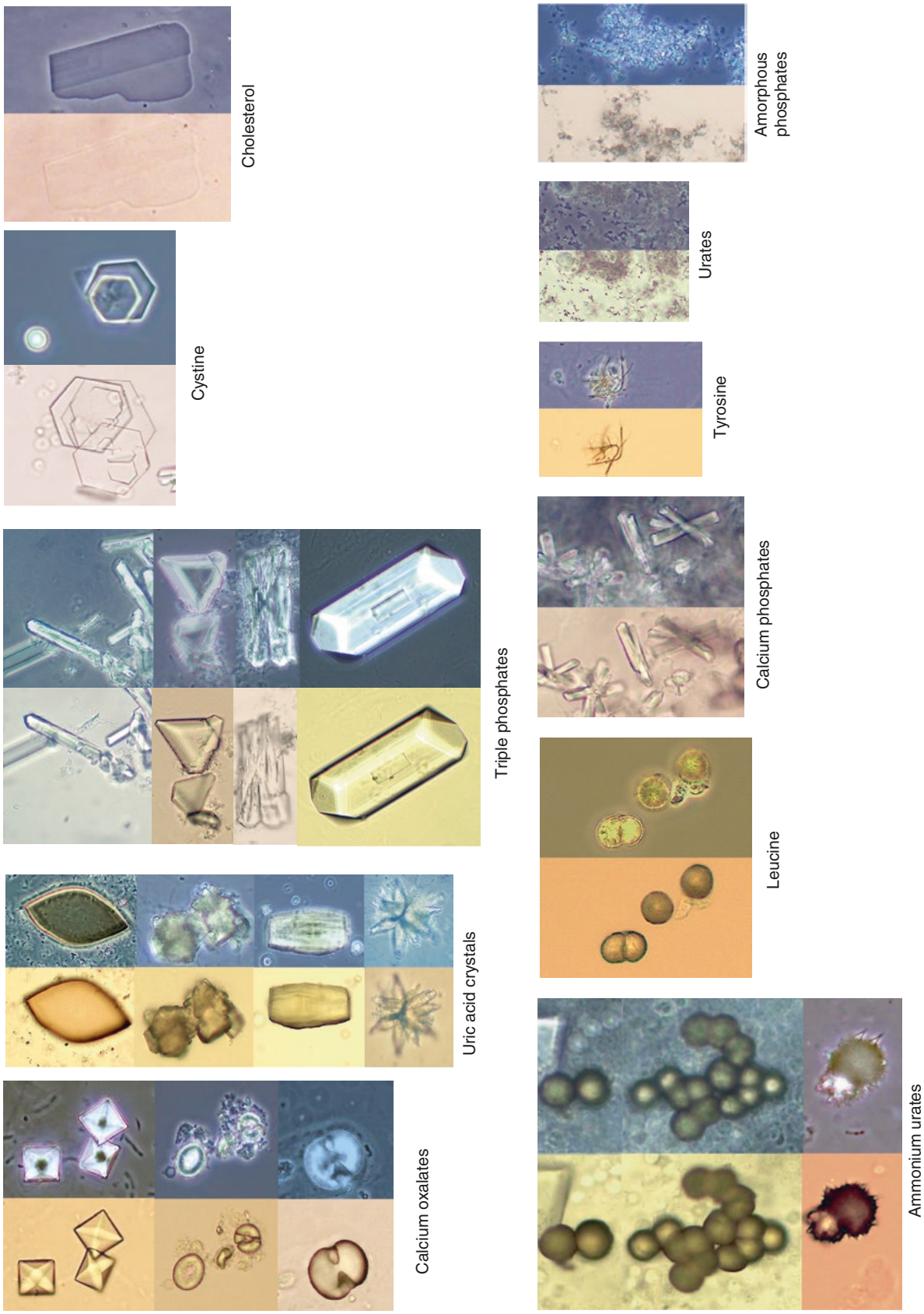


Fig. 11.83 Overview of urine crystals: *left*, bright-field and *right*, phase-contrast mode

11.11.1 Cystine

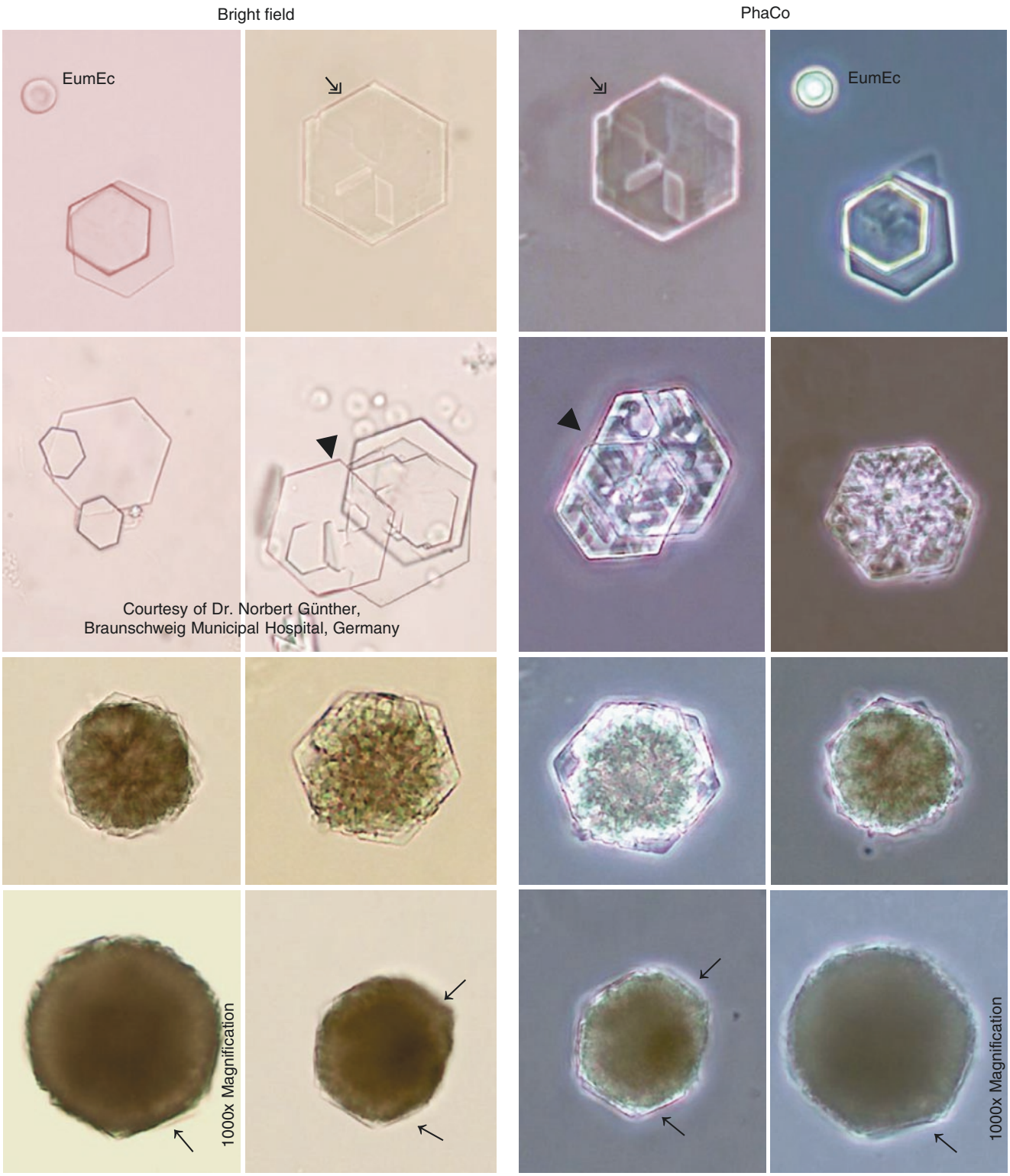


Fig. 11.84 The amino acid cystine crystallizes to hexagonal crystals in an acidic environment (pH<6). The hexagonal discs can lie individually (↘) or on top of one another (►). Cystine crystals from an older urine sample exhibit a coarse internal structure and assume a brownish color. It is sometimes no longer possible to differentiate the typical hexagonal contour (→)

11.11.2 Cholesterol

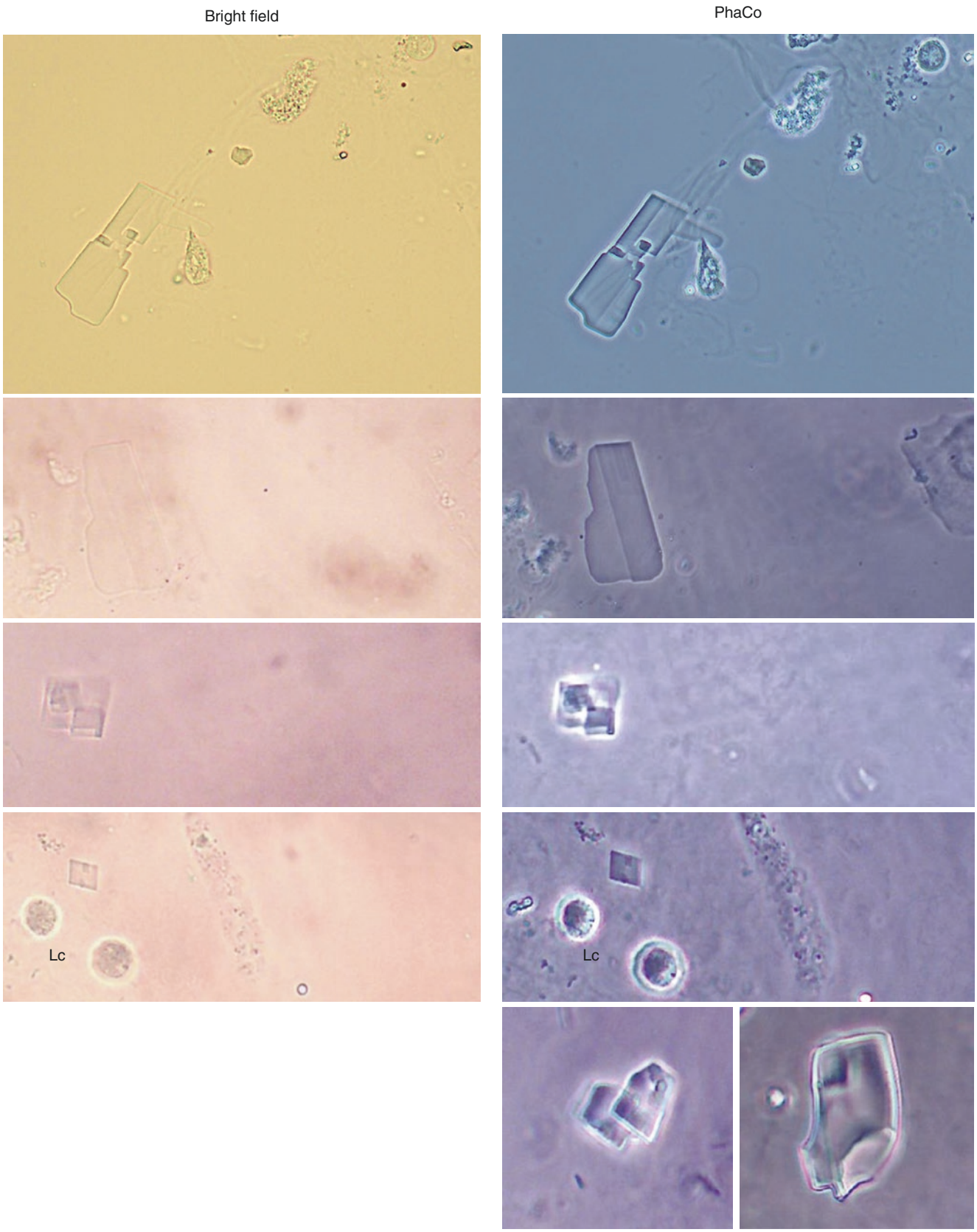


Fig. 11.85 Cholesterol must not be confused with glass fragments (e.g., from the cover glass). In bright-field mode, the aperture diaphragm on the condenser should be slightly closed in order to better identify the crystals, otherwise these fine crystalline components are easily overlooked

11.11.3 Tyrosine Crystals

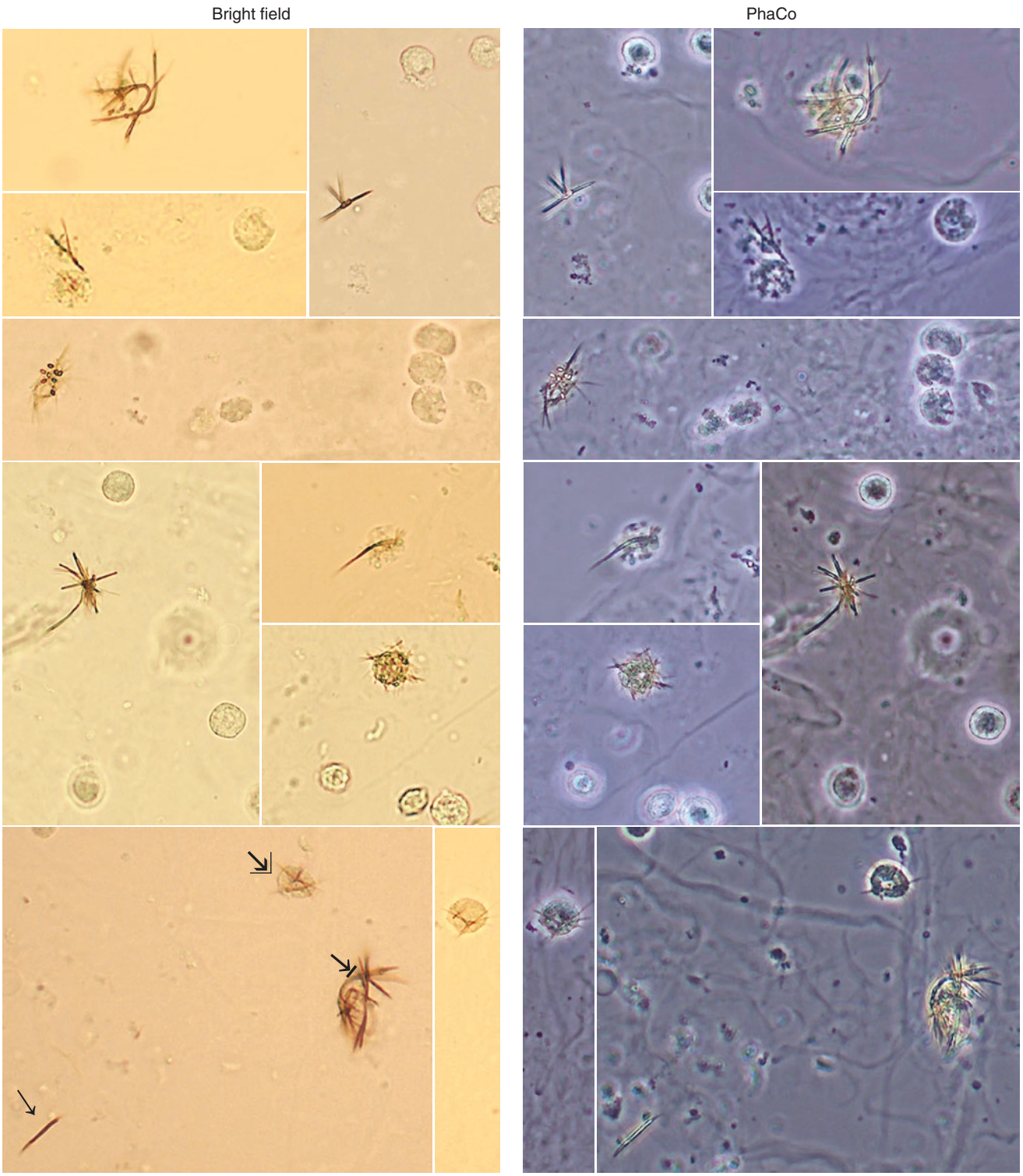


Fig. 11.86 Tyrosine crystals are an extremely rare finding in urine sediment. Tyrosine needles are found: individually (→), as (unbent or bent) needle networks (↠), and piercing leukocytes (↘) and thus in an intracellular location. Individual brownish-yellow tyrosine needles are more difficult to see in PhaCo than in bright-field mode

11.11.4 Comparison: Leucine–Ammonium Urates

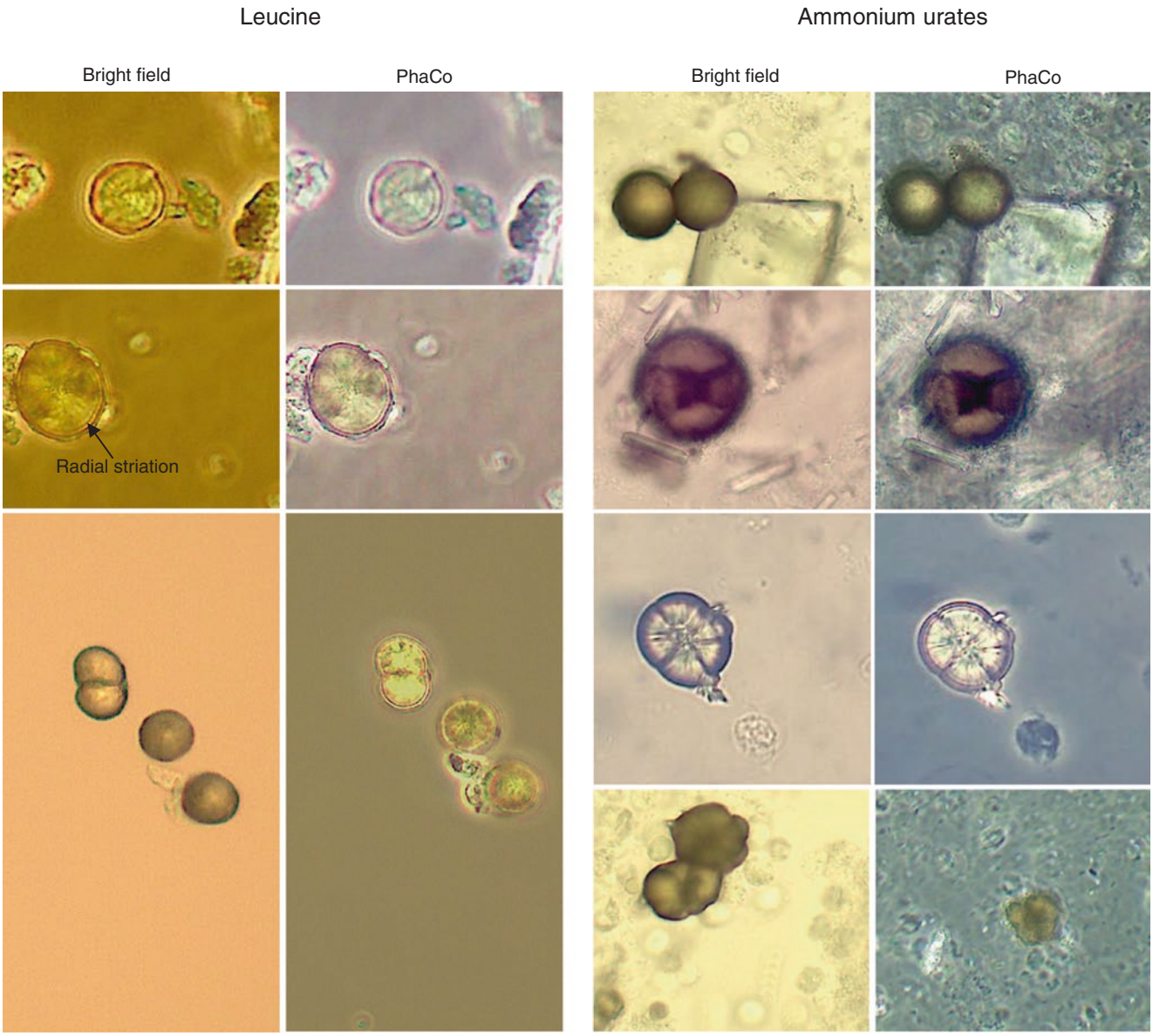


Fig. 11.87 Ammonium urates are easily confused with the extremely rare leucine or 2,8-dihydroxyadenine and xanthine crystals [see Hesse (2009)]. Paying attention to the microscopic environment is recommended for better differentiation: ammonium urates are formed by urease-positive bacteria as part of an inflammatory reaction. Rarely occurring crystals such as 2,8-dihydroxyadenine and xanthine can only be clearly differentiated using infrared spectroscopy

11.11.5 Ammonium Urates

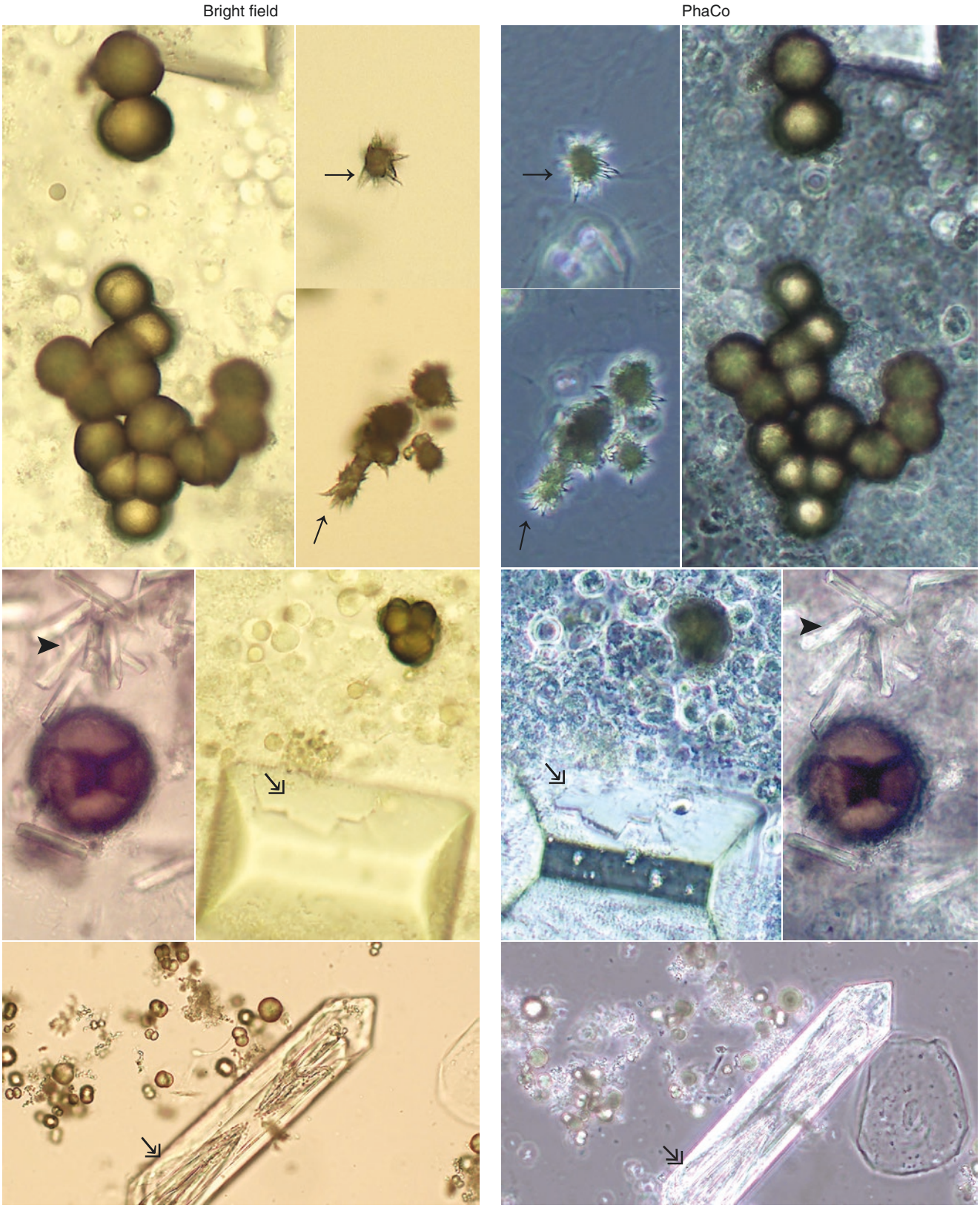


Fig. 11.88 Brown ammonium urate spheres—some with thorn-like projections (→)—can be particularly well differentiated in bright-field mode. The spheres lie individually or in a conglomerate—and can also occur together with triple phosphates (⇲) and calcium phosphates (➤) in the case of bacterial urinary tract infection with urease-positive bacteria

11.11.6 Calcium Oxalates

The size of calcium oxalates varies greatly. They are found individually or in conglomerates. Calcium oxalates crystallize in a wide variety of shapes. The most common shapes are angular (envelope-shape) (→), round (↘) , oval (➤), and hourglass-shaped (➢). Round calcium oxalate shapes must not be confused with eumorphic erythrocytes (☞).

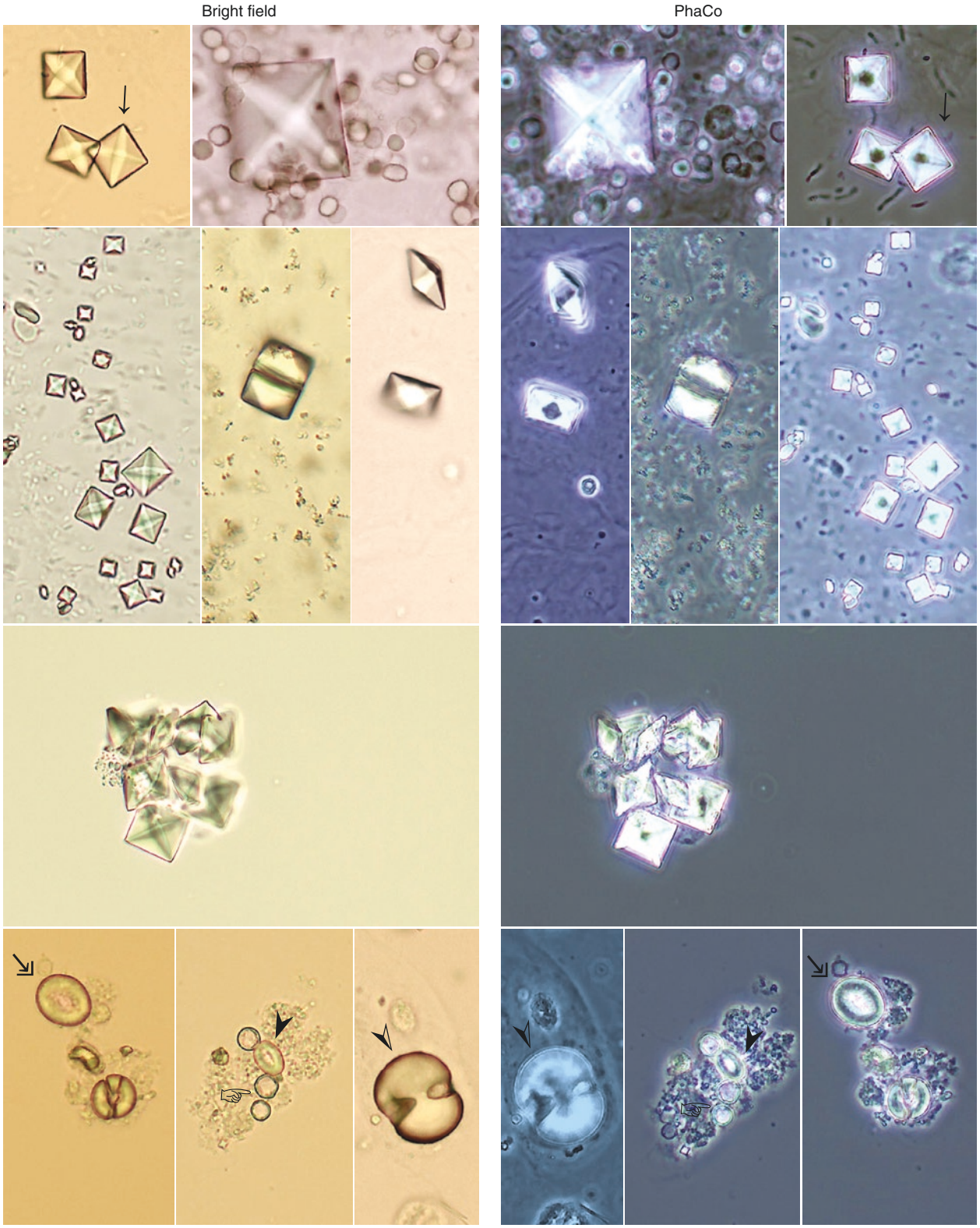


Fig. 11.89 Ca-oxalates: angular, envelope-shape, round, oval, hourglass-shape

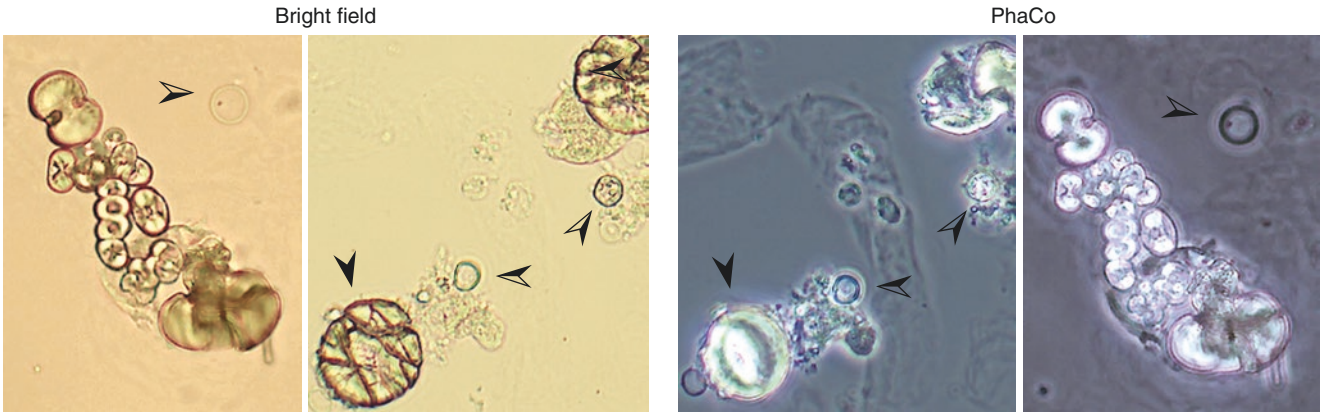


Fig. 11.90 Roundish-oval as well as hourglass-shaped Ca-oxalates and eumorphic erythrocytes (➢). Large crystals can break up spontaneously (➤)

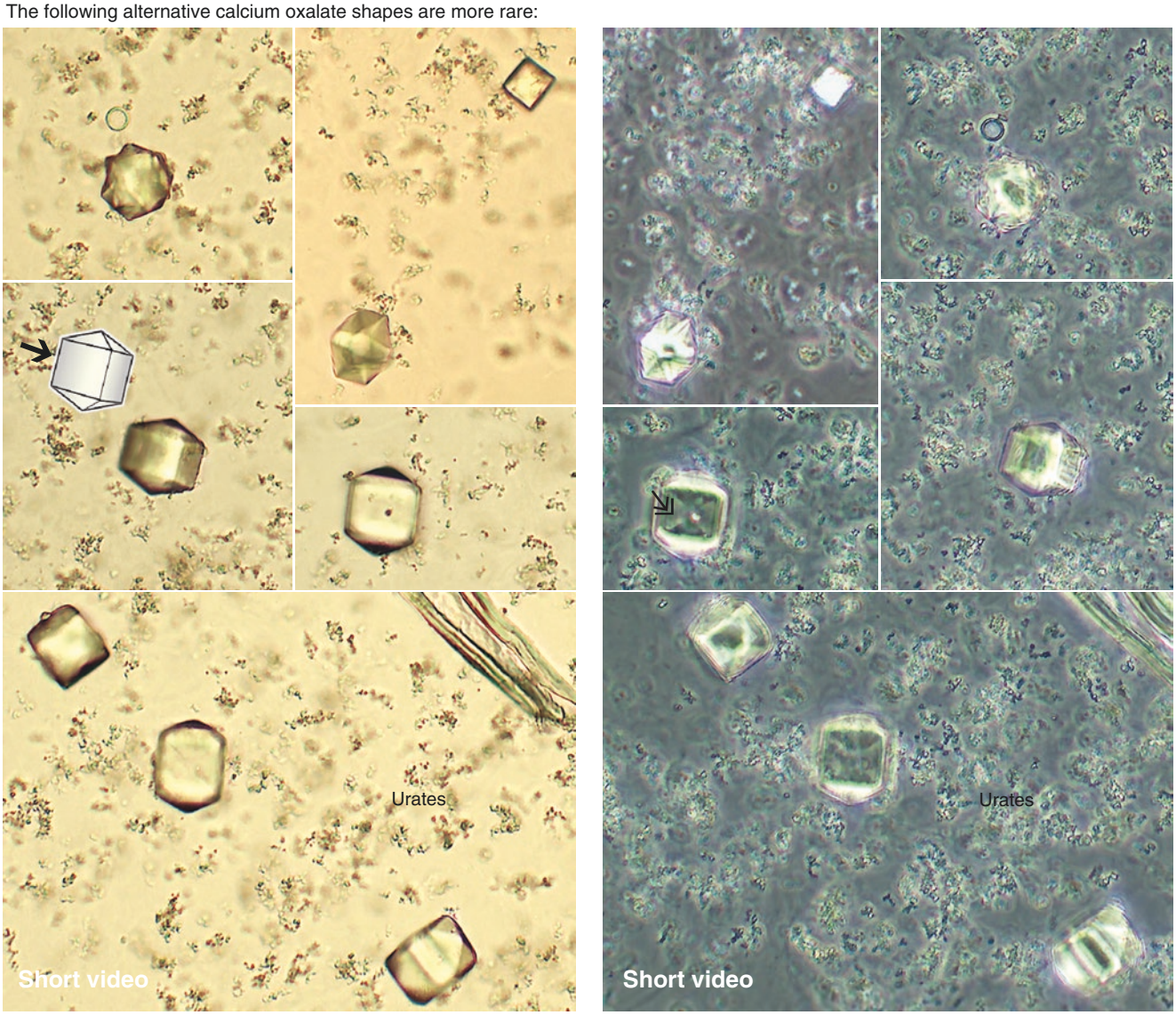


Fig. 11.91 Rare crystalline Ca-oxalate shapes. The three-dimensional structure needs to be taken into account with this crystallization shape (see schematic illustration ➔). The contour of this geometric shape resembles a hexagonal cystine crystal and should not be confused with it! Sometimes one can see the typical cross (⇲) inside these Ca-oxalate forms, as in the envelope-shape, using the phase-contrast technique. (see Videos 11.18 and 11.19)

11.11.7 Uric Acid Crystals

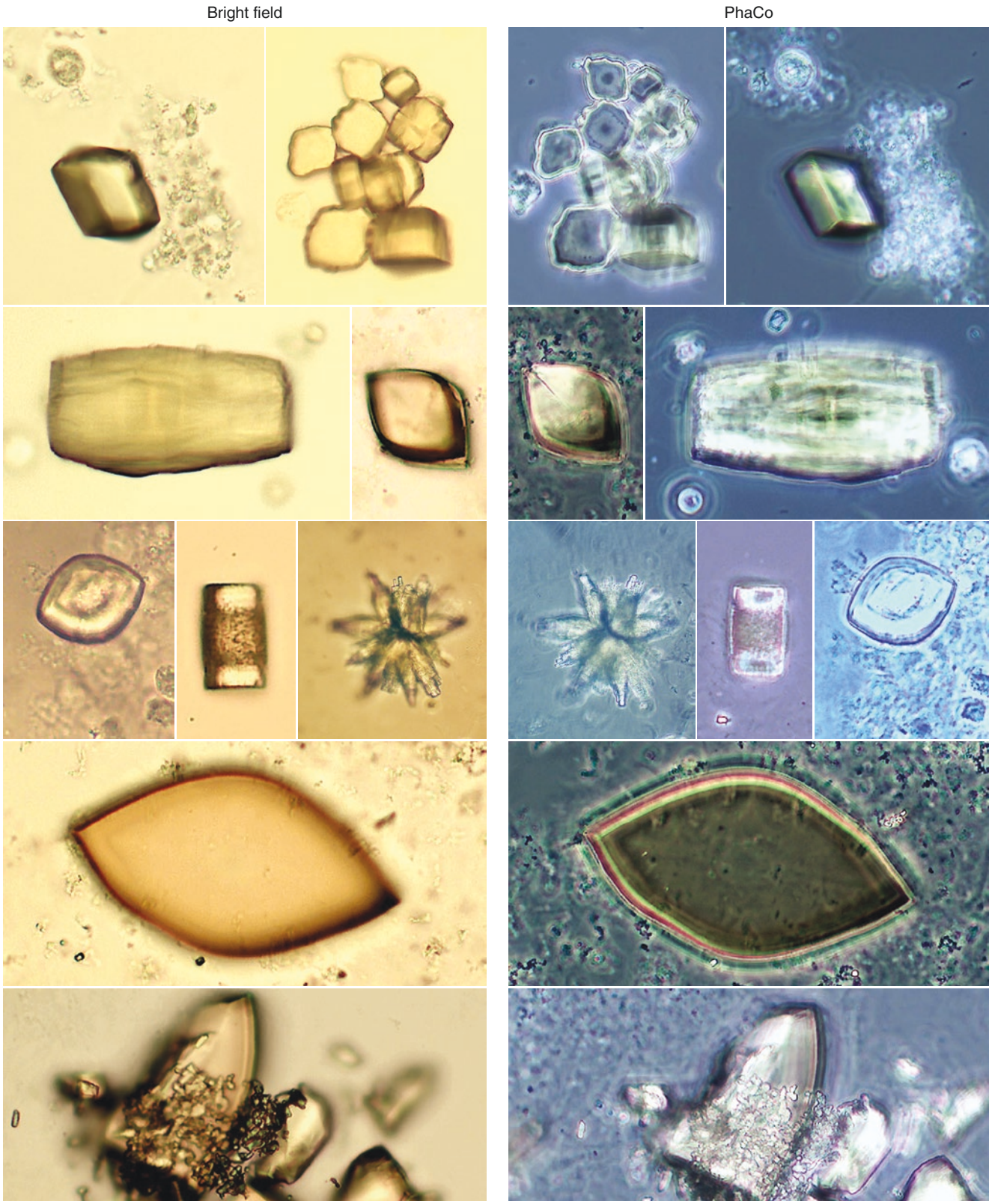


Fig. 11.92 The typical intrinsic color of the crystals is better visualized in bright-field than in phase-contrast mode. Uric acid crystals can crystallize in highly varying shapes and sizes (barrel, diamond, and rosette shape)

11.11.8 Urates: Semi-quantitative Analysis

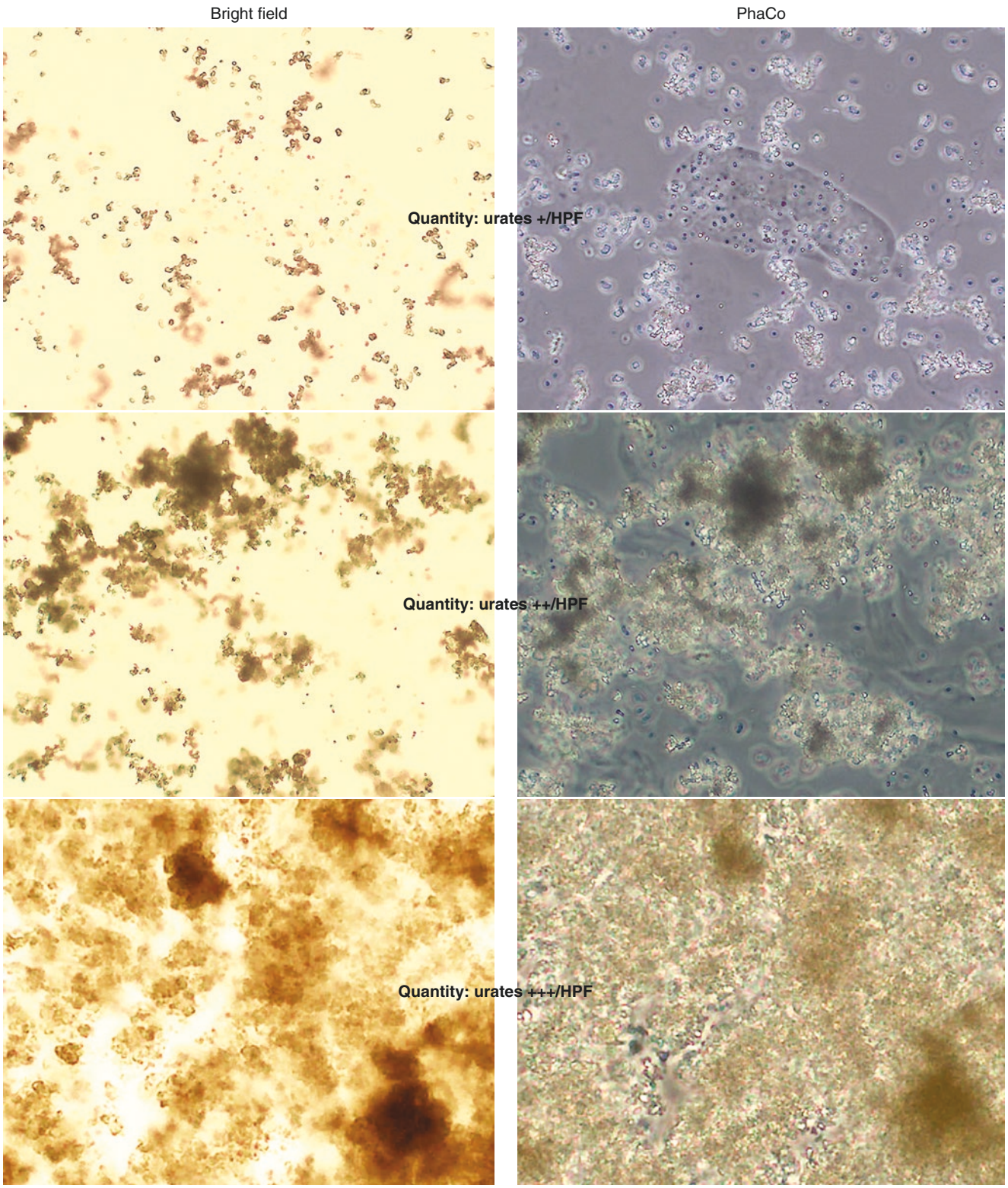


Fig. 11.93 If there is a high concentration of urates in urine, a brownish-red "brick dust sediment" can be seen macroscopically. The color of the urates can be reliably determined microscopically in bright-field mode. Urine pH < 6

11.11.9 Amorphous Phosphates (Tricalcium and Trimagnesium Phosphates)

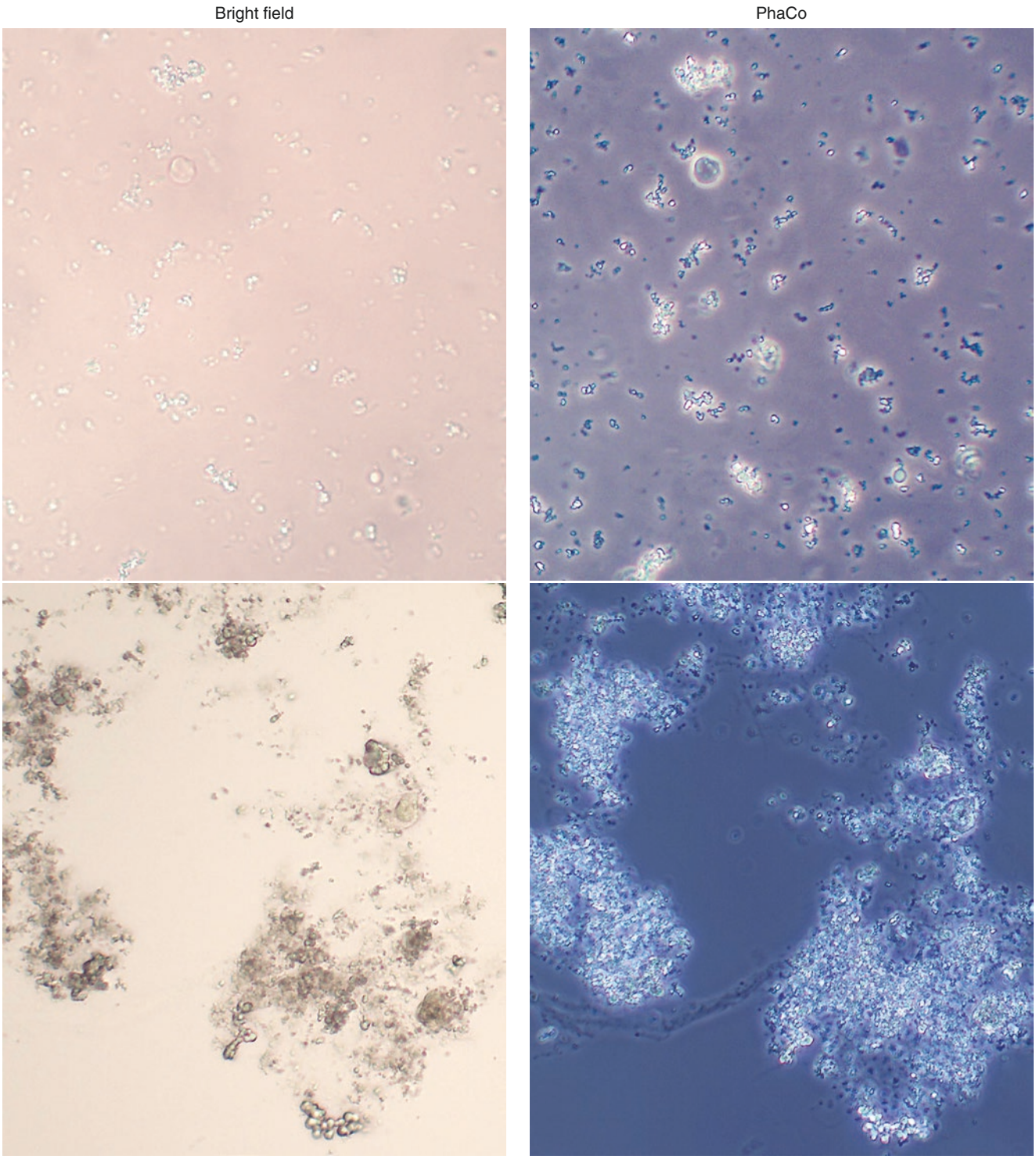


Fig. 11.94 Amorphous phosphates crystallize in alkaline urine as colorless (see bright-field), irregularly shaped granules the size of sand grains. In the case of an increased presence of these crystals, a gray-white sediment can be seen macroscopically

11.11.10 Comparison: Urates–Amorphous Phosphates

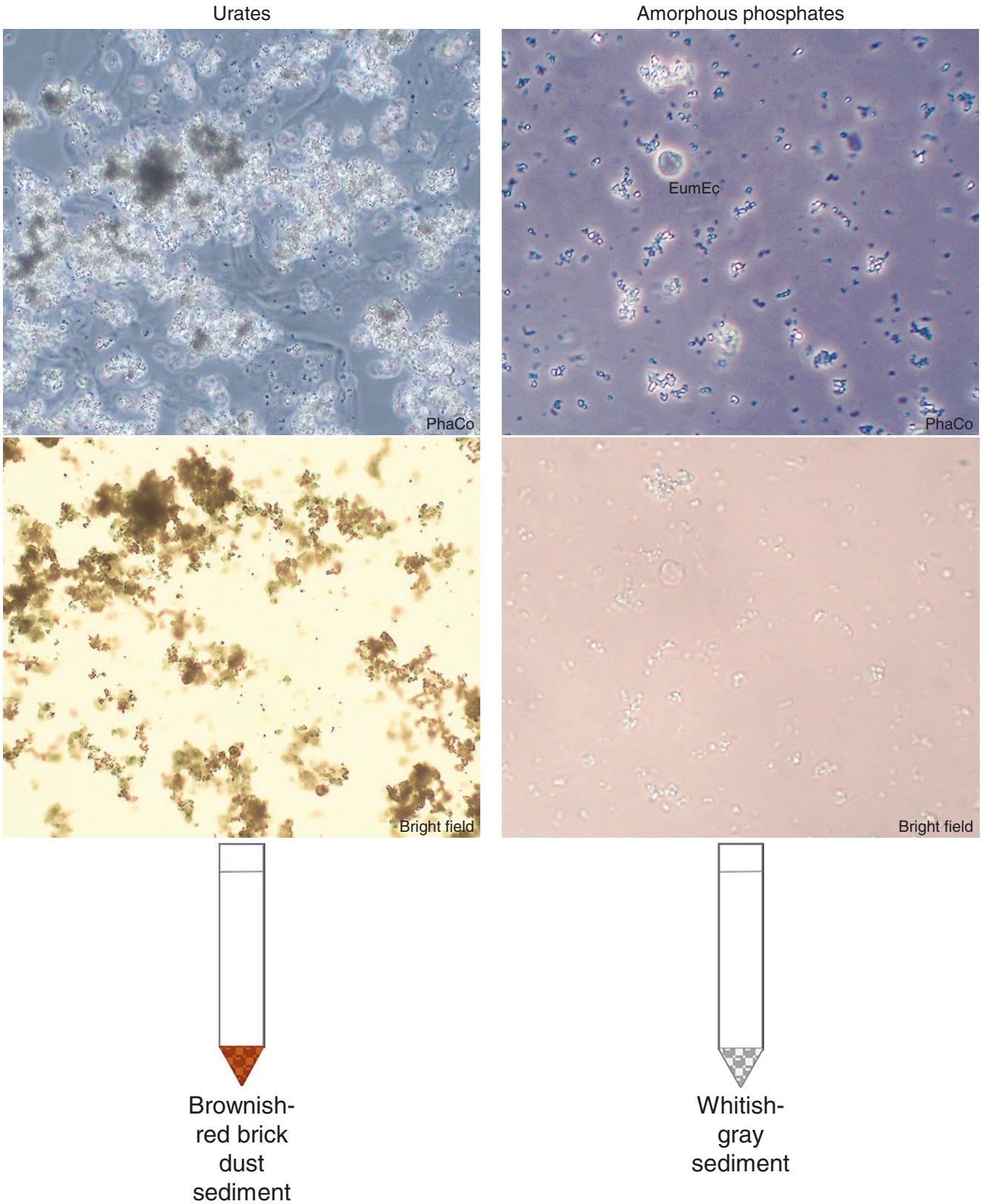


Fig. 11.95 Comparison: urates–amorphous phosphates

11.11.11 Triple Phosphates

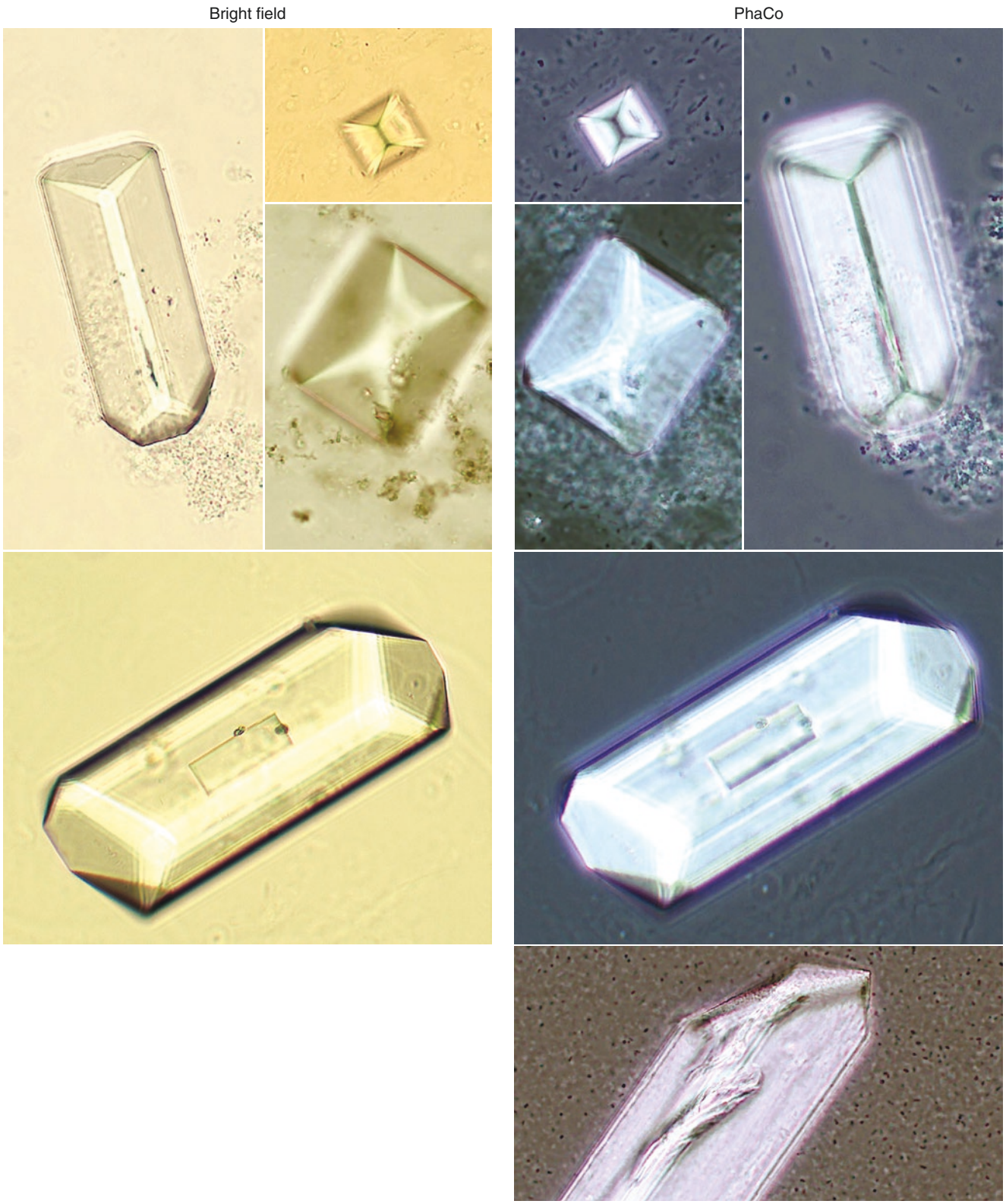


Fig. 11.96 Coffin-lid shape. The size of triple phosphates varies considerably. Typical concomitant findings include an alkaline pH value (urine pH 8) and bacteriuria. In the case of concomitant leukocyturia, the suspicion of a bacterial urinary tract infection rises

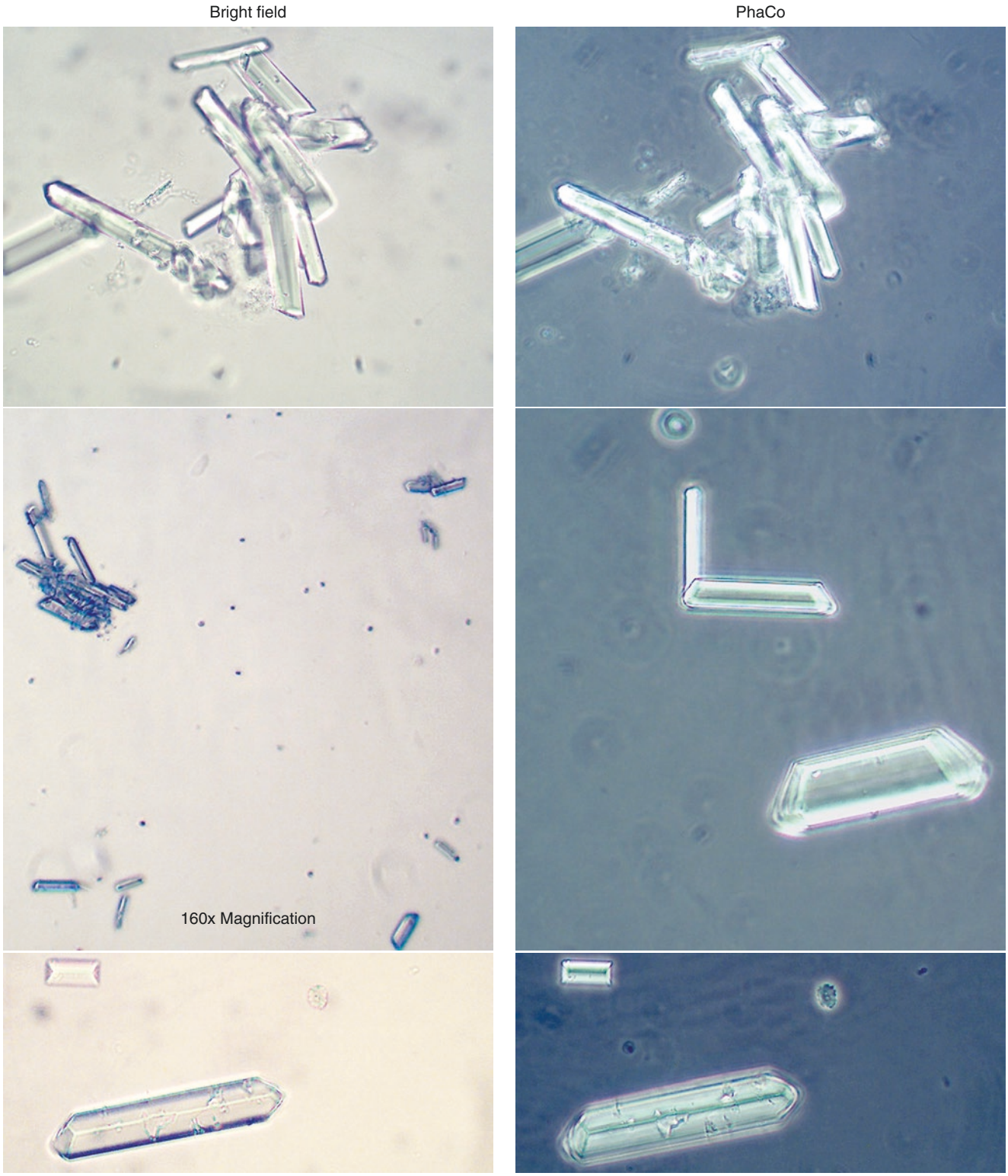


Fig. 11.97 Elongated form of triple phosphates, partially in clusters

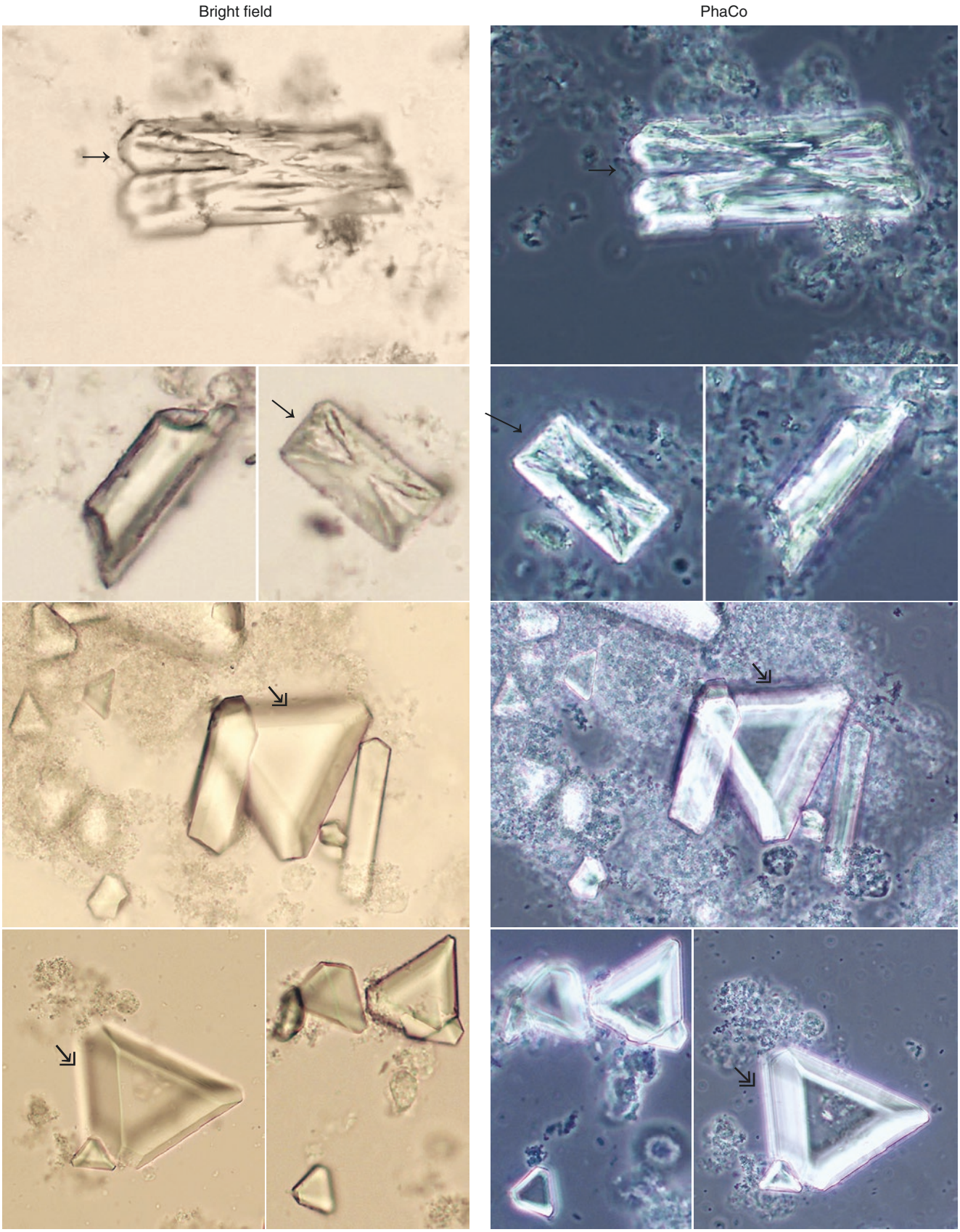


Fig. 11.98 Rare triple phosphate shapes: fern-leaf shape (→) and triangular (↘)

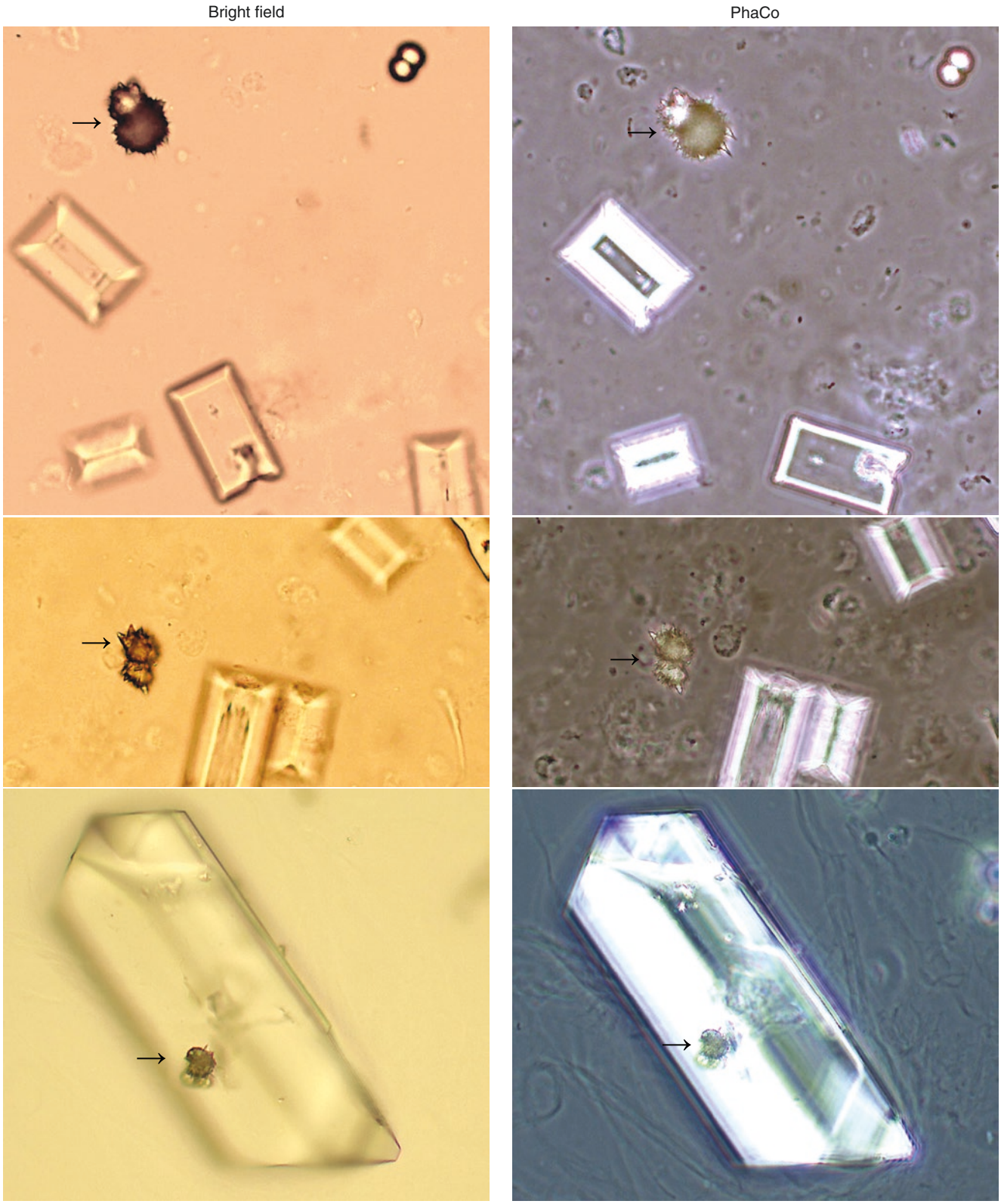


Fig. 11.99 Triple phosphate and ammonium urate crystals (→) are formed by the metabolization of urease-positive bacteria. In addition to the triple phosphates, one can also clearly see the brown ammonium spheres in bright-field mode

11.11.12 Calcium Phosphates

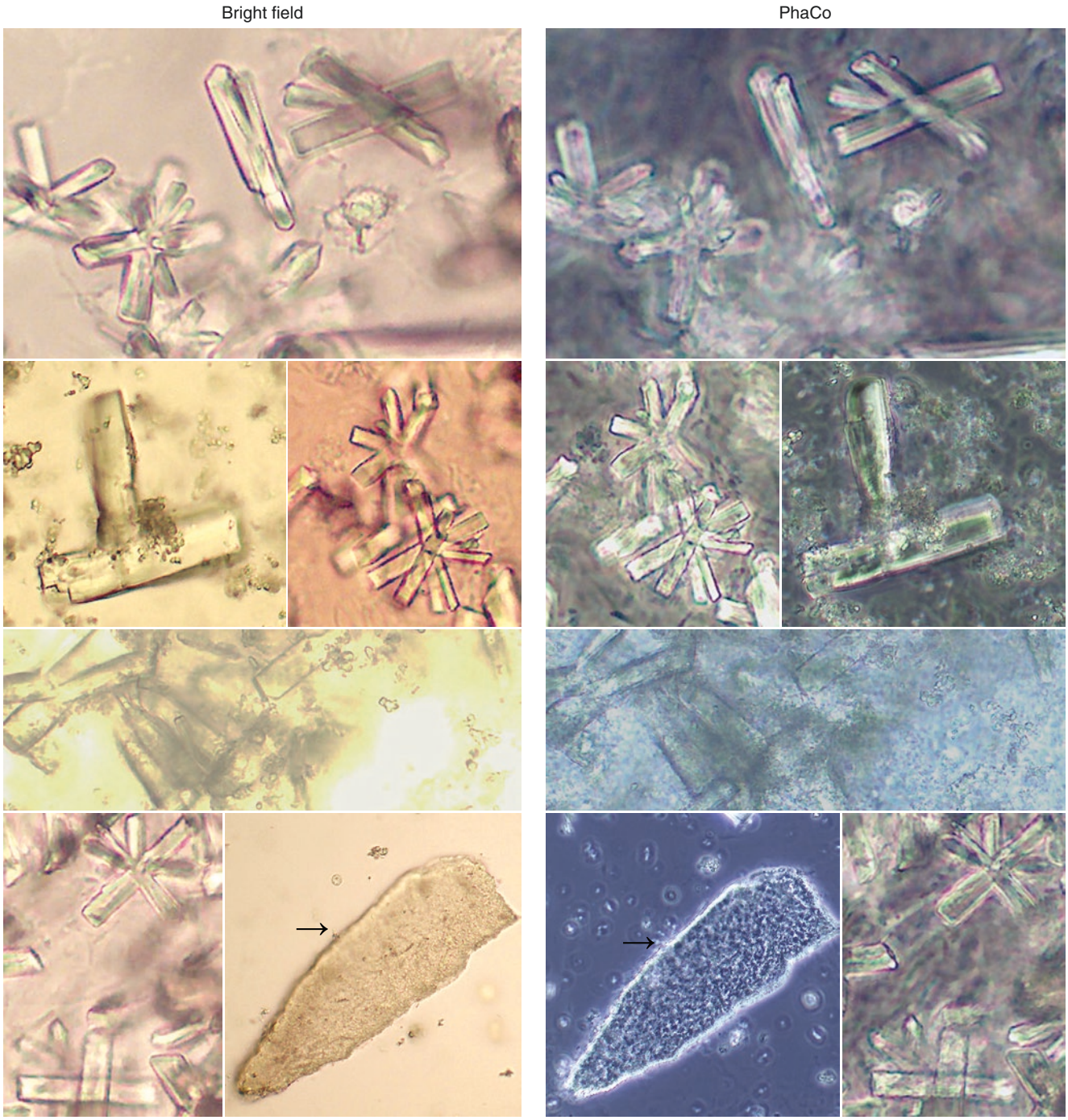


Fig. 11.100 The fan-shaped arrangement of the rectangular or wedge-shaped crystals is typical. Rarely, calcium phosphate also crystallizes in plate form with irregular corners (→) in alkaline to slightly acidic urine

11.11.13 Drug Crystals

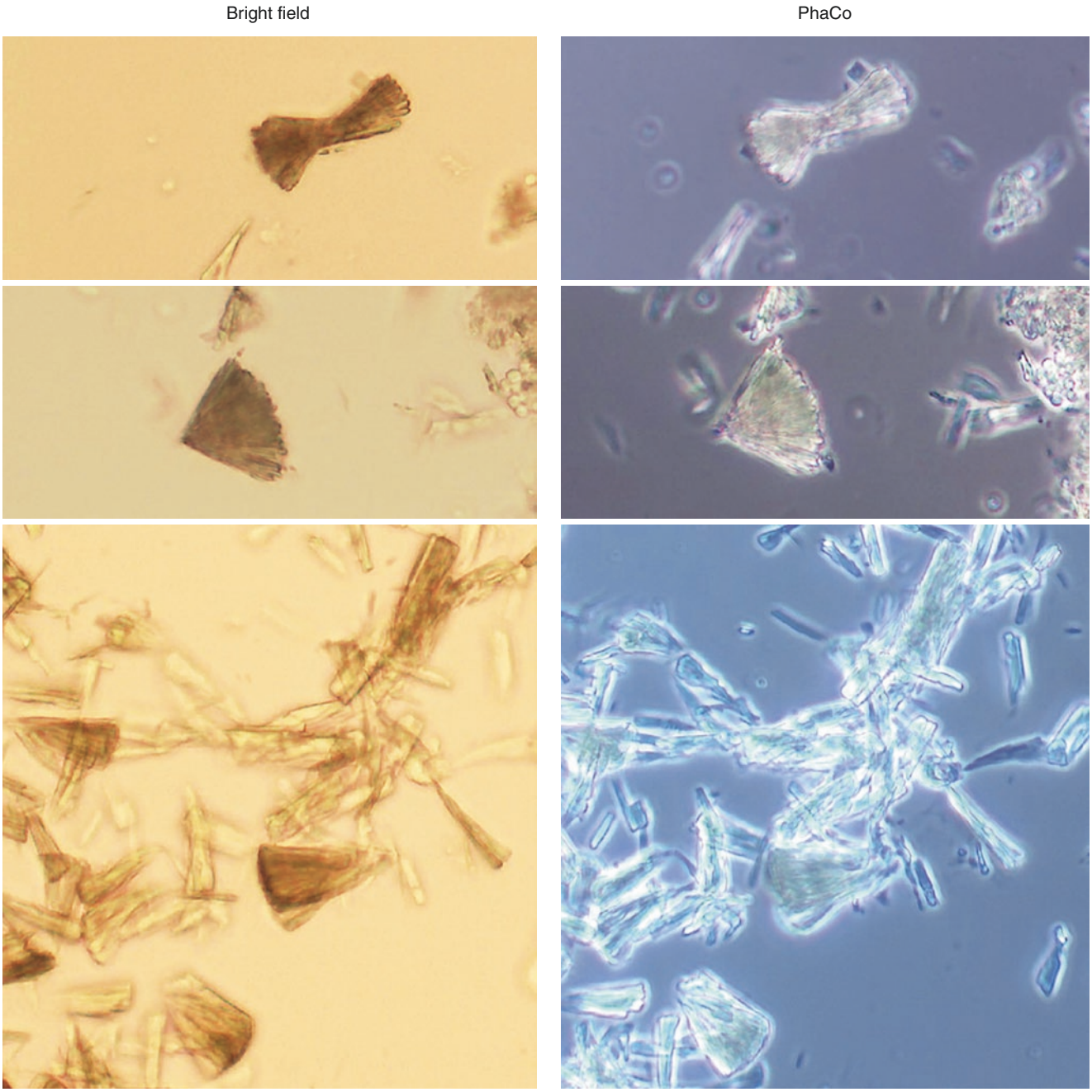


Fig. 11.101 Crystalline excretions of drugs in urine often exhibit striking but atypical crystallization shapes. These images could be of amoxicillin crystals

11.12 Artifacts

11.12.1 Glass Fragments

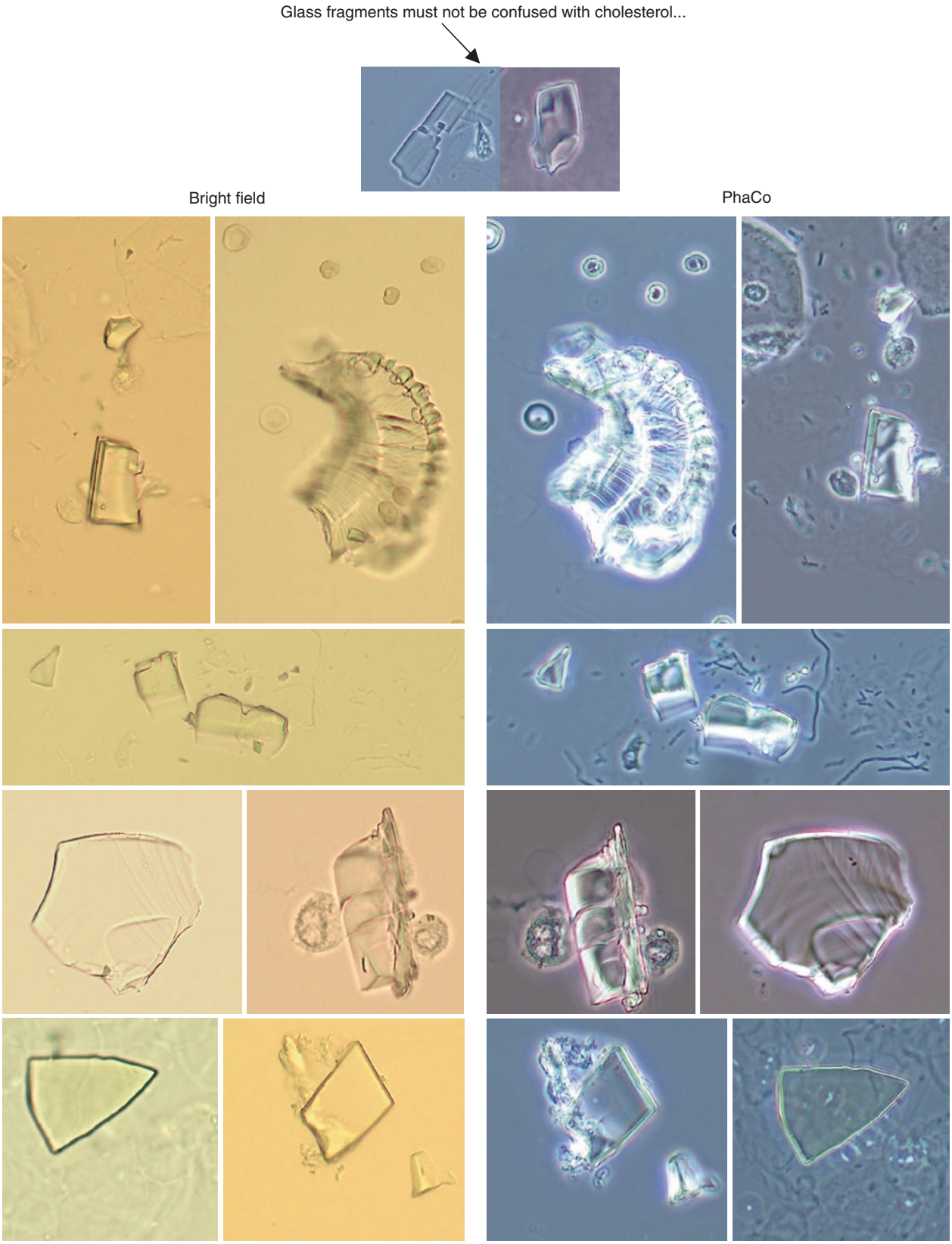


Fig. 11.102 Glass fragments

11.12.2 Pollen

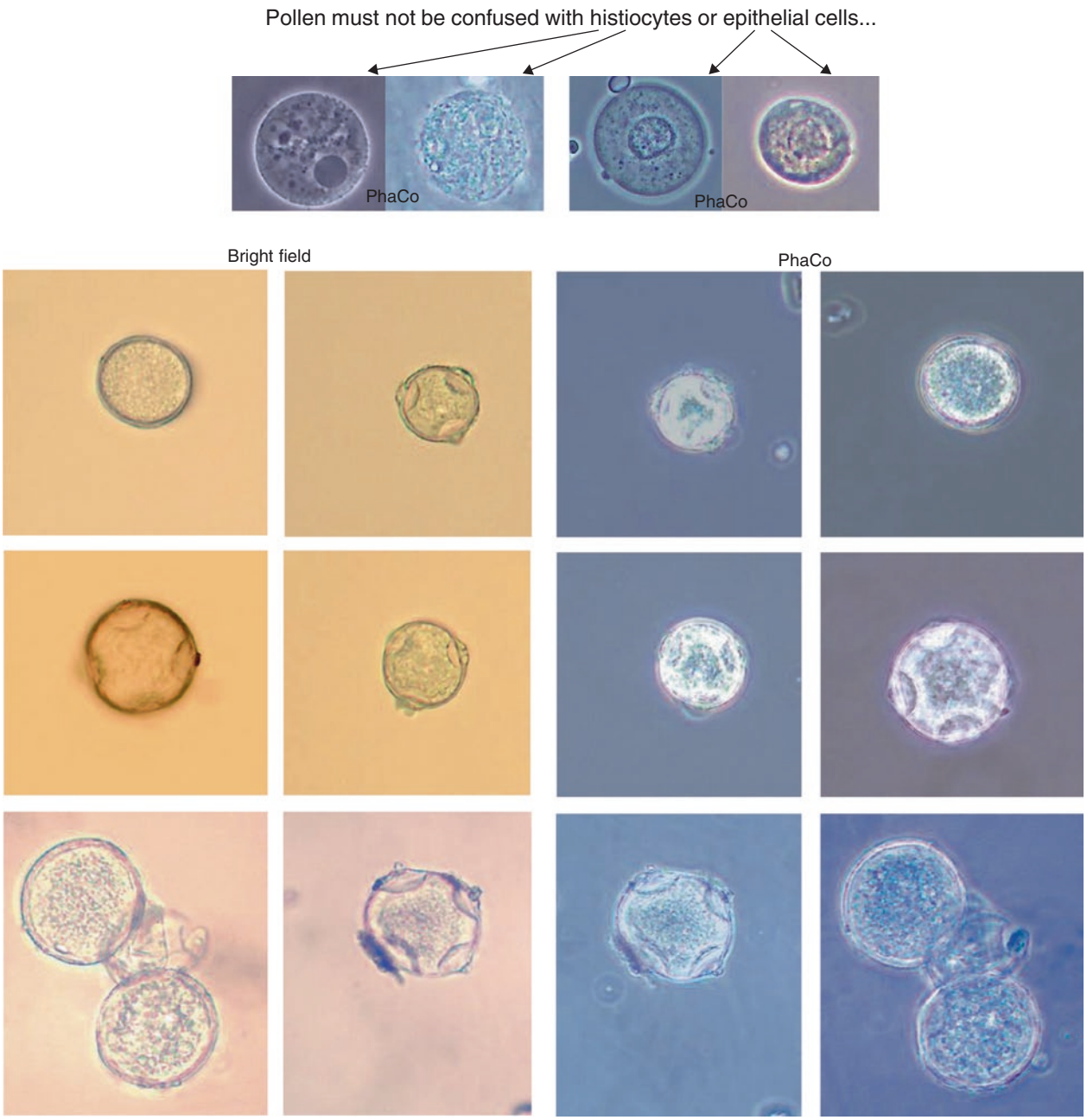


Fig. 11.103 Pollen

11.12.3 Starch Grains

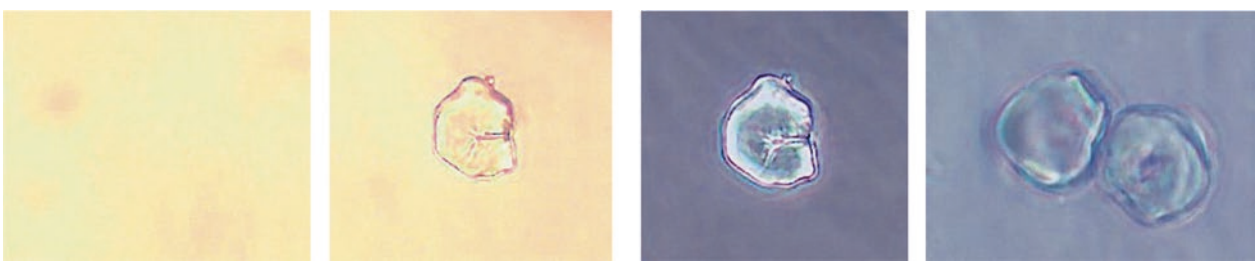


Fig. 11.104 Starch grains

11.12.4 Cylindrical Artifacts

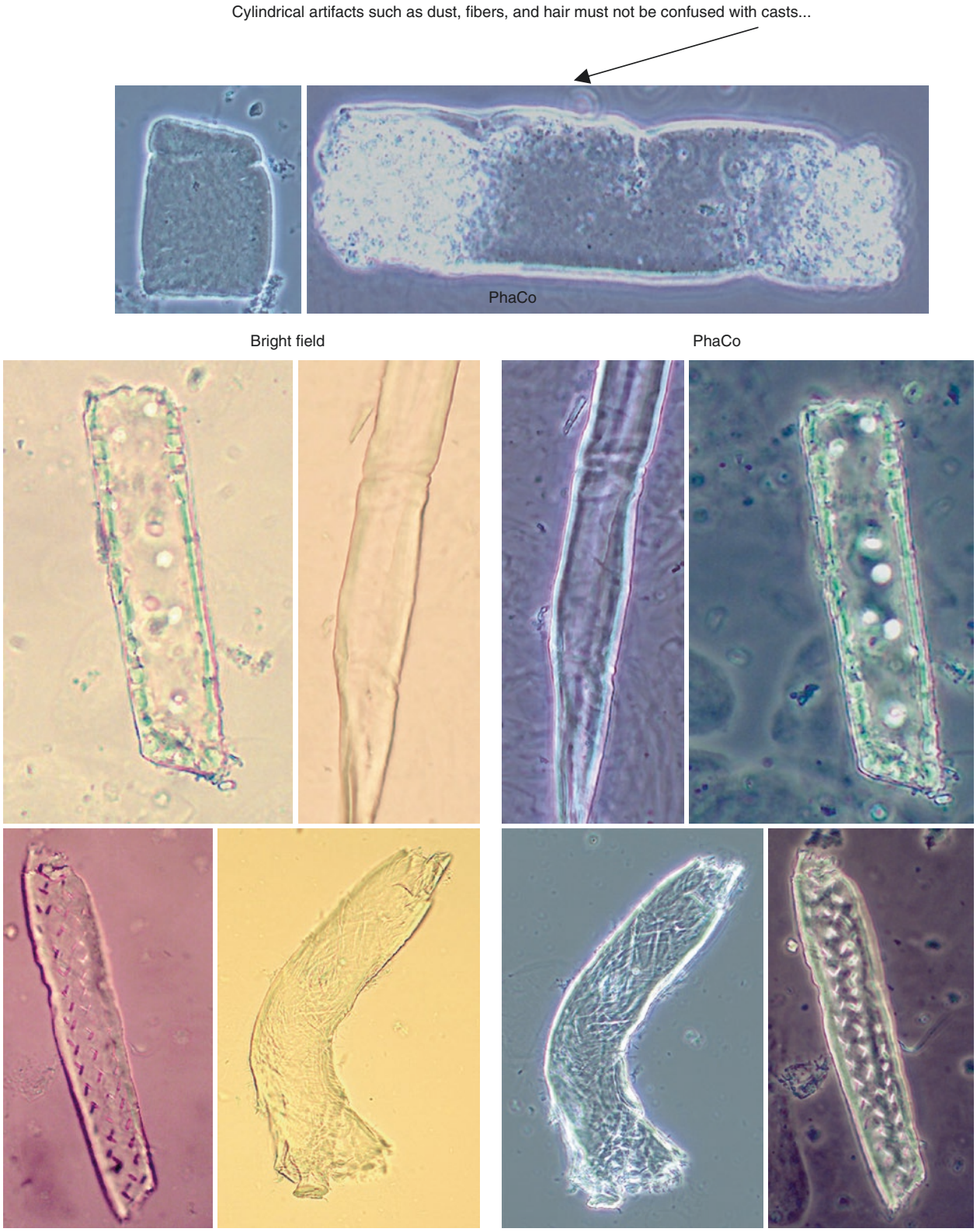


Fig. 11.105 Cylindrical artifacts I

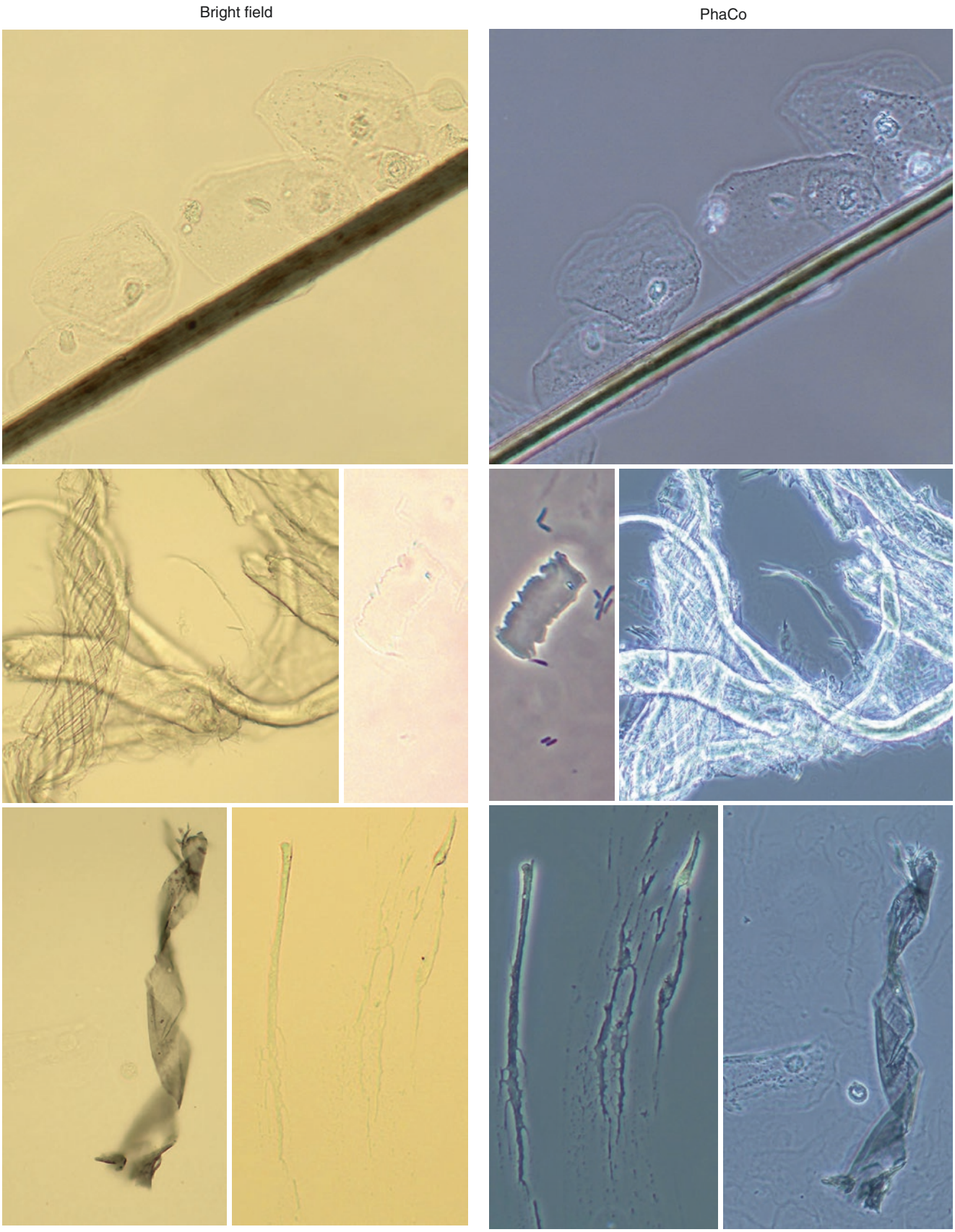


Fig. 11.106 Cylindrical artifacts II

11.12.5 Air Bubbles and Fat Droplets

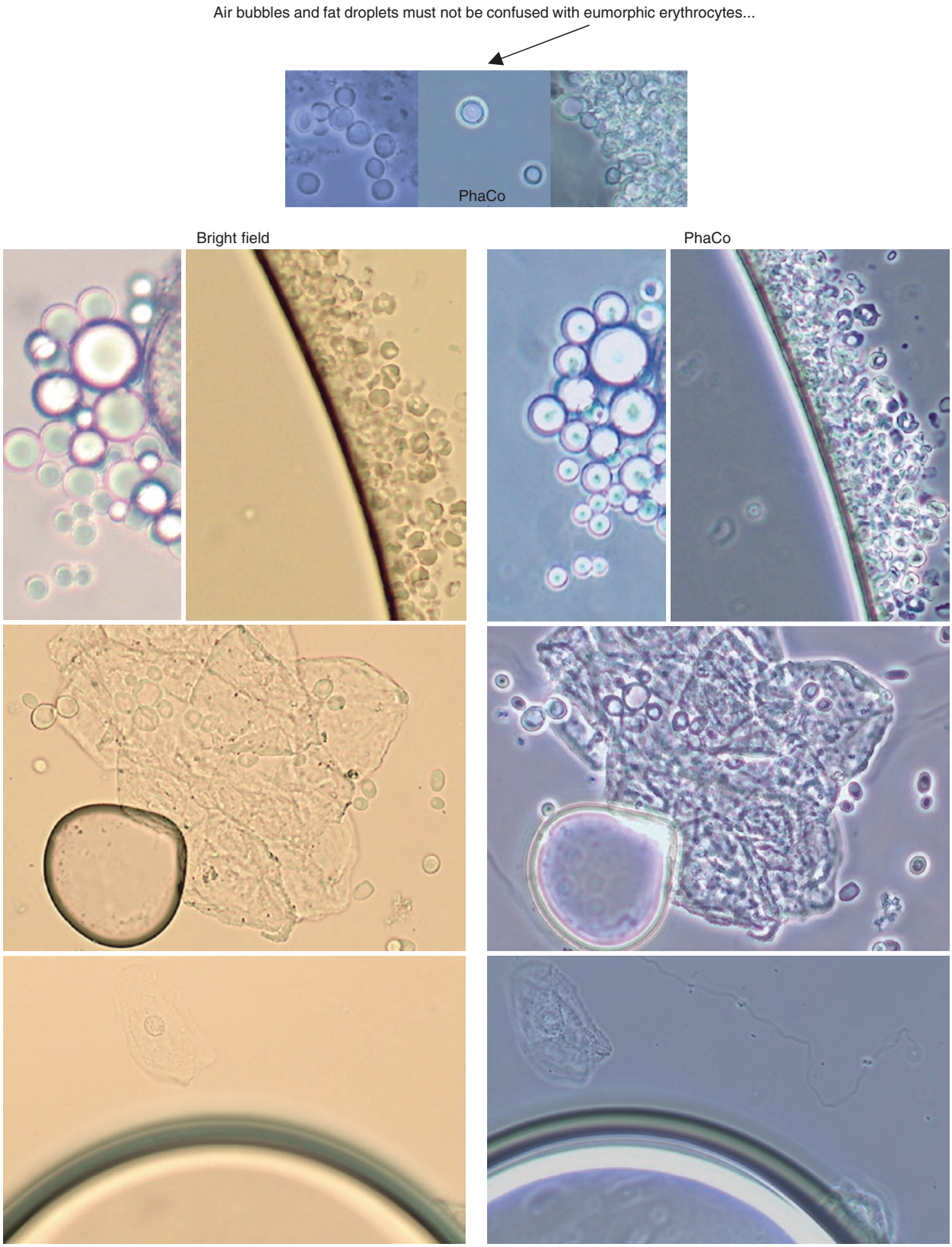


Fig. 11.107 Large and small air bubbles

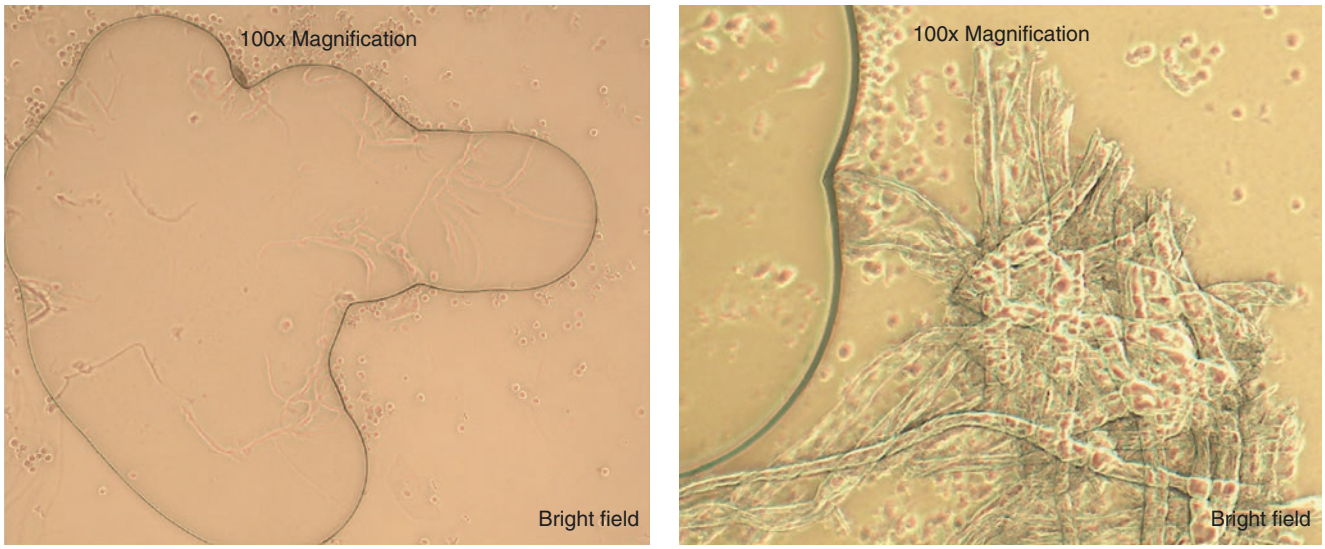


Fig. 11.108 Large air bubbles at 100× magnification

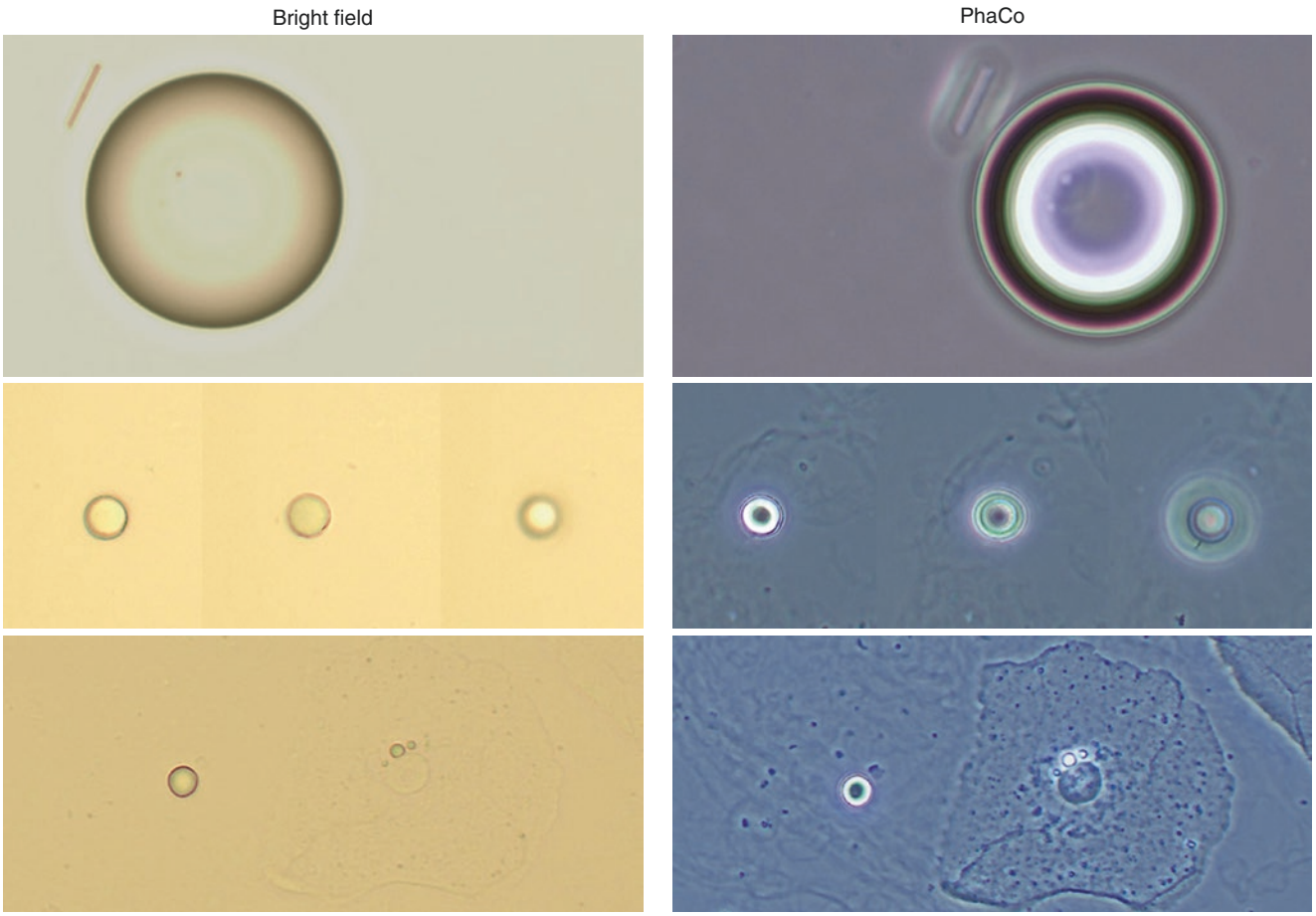


Fig. 11.109 Fat droplets: Turning the micrometer knob causes the fat droplet to alter its luminosity significantly

11.12.6 Other Artifacts

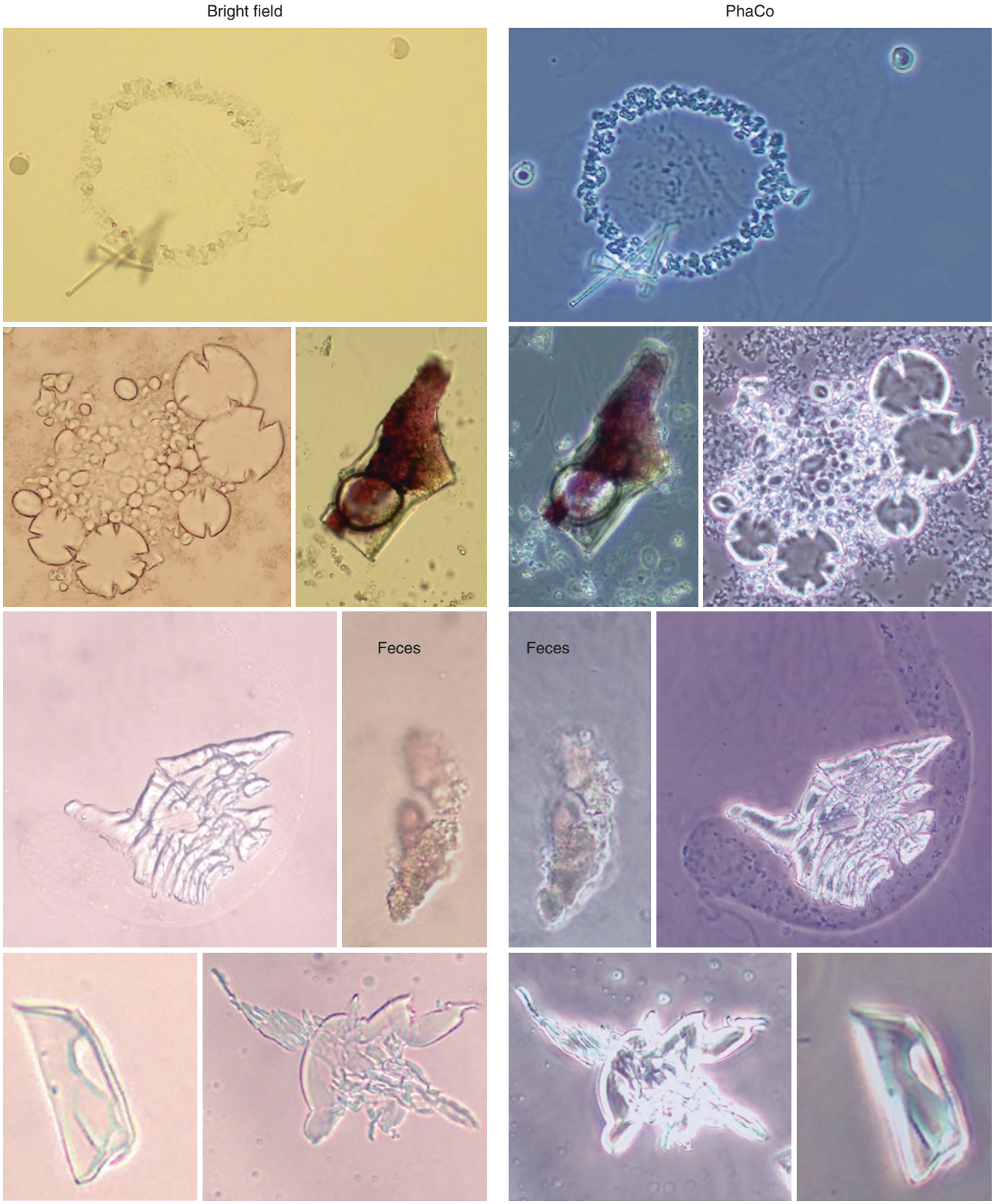


Fig. 11.110 Other artifacts

Part III

Microscopic Urine Sediment: Analysis and Findings

12

12.1 Introduction to the Analysis and Diagnosis of the Microscopic Urinary Sediment Image

The following enables the reader to practice the analysis and hence the classification and semi-quantitative recording of urinary sediment constituents per high power field.

- If the microscopic analysis of urine sediment reveals a similar distribution of urinary sediment constituents in all high power fields (HPF) as in the respective figure, one can perform analysis as described below.
- Particular attention should be paid to erythrocytes, leukocytes, and bacteria. Erythrocytes are differentiated into eumorphic and dysmorphic erythrocytes and acanthocytes and their percentage determined.
- In order to better assess the quality of the urine sample, one counts squamous epithelial cells per HPF. An increased presence of these epithelial cells generally indicates that the sample is not taken from midstream urine.
- In addition, all other urinary sediment constituents per HPF are recorded semi-quantitatively. Casts are an exception. Here, one adds the respective cast types in all analyzed HPF (usually 20) and gives the respective total of the relevant cast type as the result.
- Artifacts and mucus threads need to be identified, but not mentioned in the subsequent findings.
- For a discussion on whether semi-quantitative determination is carried using numerical values or with the aid of crosses, the reader is referred to Chap. 5, Sect. 5.8, "Semi-quantitative Analysis/Units".
- In order to better view and classify urinary sediment constituents in viscous and sometimes highly cell-rich urine sediment samples, microscopy analysis is performed on a narrow area or at the specimen border.

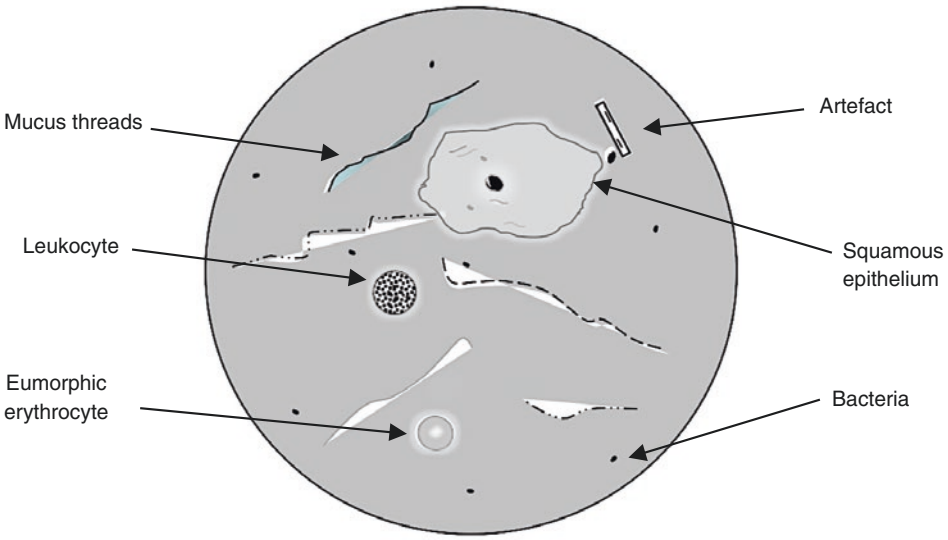
The basics of correct analysis and diagnosis of the microscopic urinary sediment image can be practiced using the following typical schematic examples of diagnosis.

J. Neuendorf, *Urine Sediment*, https://doi.org/10.1007/978-3-030-15911-5_12

12.2 Illustrated Diagnostic Examples

12.2.1 Normal Findings

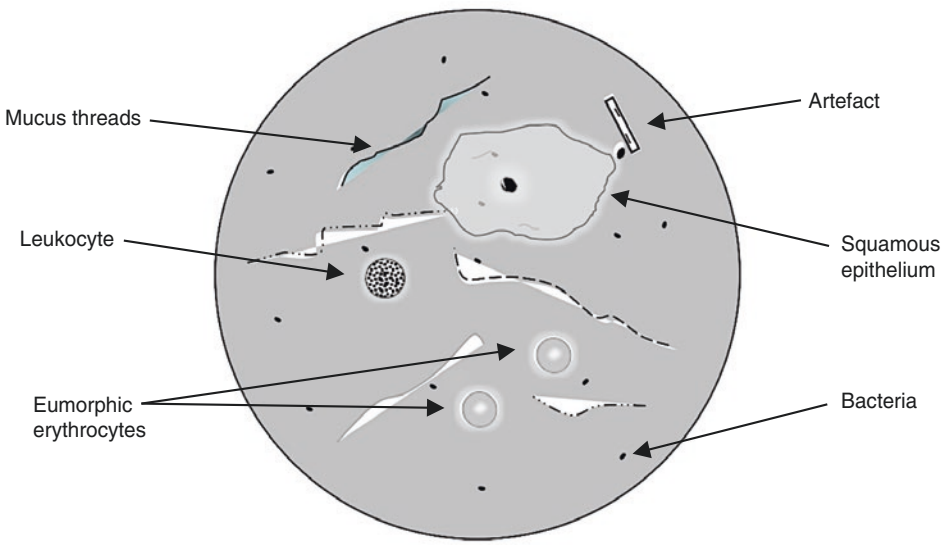
Fig. 12.1 Normal findings



EumEc:	0-1	/HPF	SqEc:	0-1	/HPF
DysEc:	-	/HPF	Bact:	(+)	/HPF
Lc:	0-1	/HPF			

12.2.2 Eumorphic Hematuria I

Fig. 12.2 Eumorphic hematuria I



EumEc:	1-4	/HPF	SqEc:	0-1	/HPF
DysEc:	-	/HPF	Bact:	(+)	/HPF
Lc:	0-1	/HPF			

12.2.3 Eumorphic Hematuria II

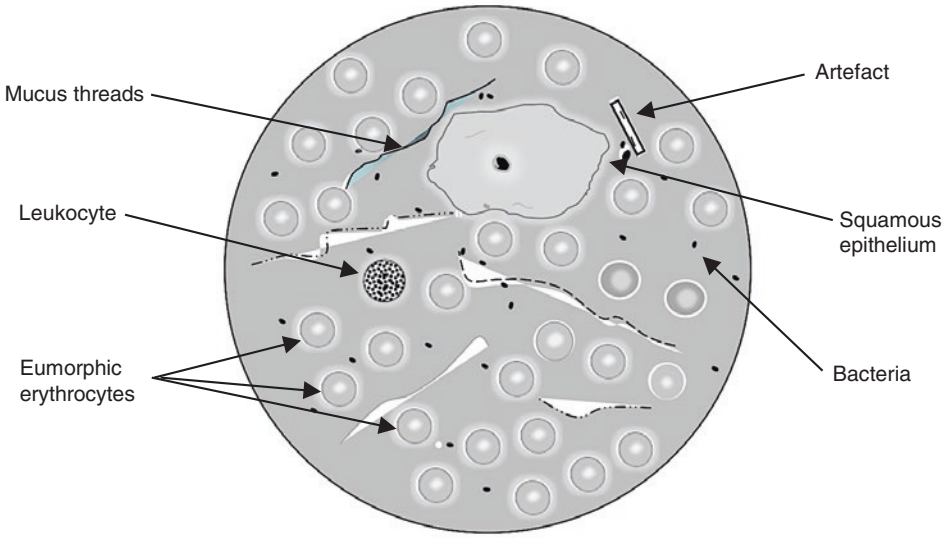


Fig. 12.3 Eumorphic hematuria II

EumEc: 15-50	/HPF	SqEc: 0-1	/HPF
DysEc: -	/HPF	Bact: (+)	/HPF
Lc: 0-1	/HPF		

12.2.4 Dysmorphic Hematuria

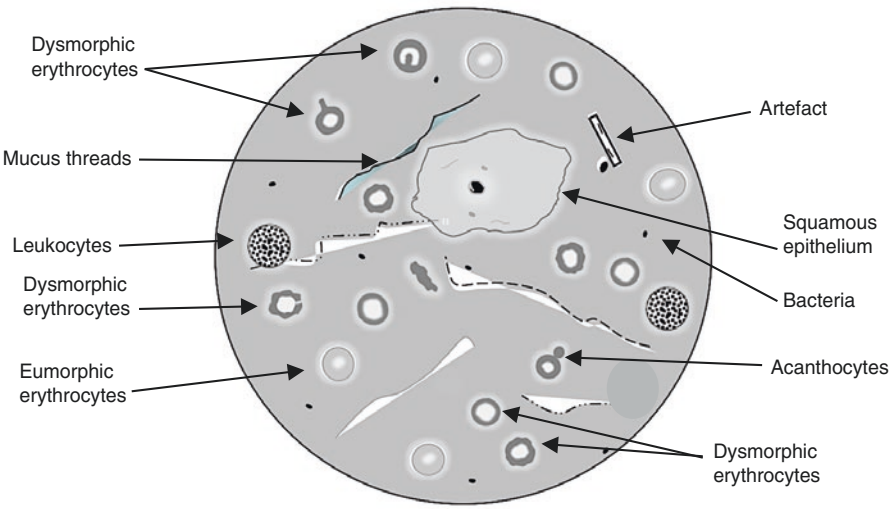
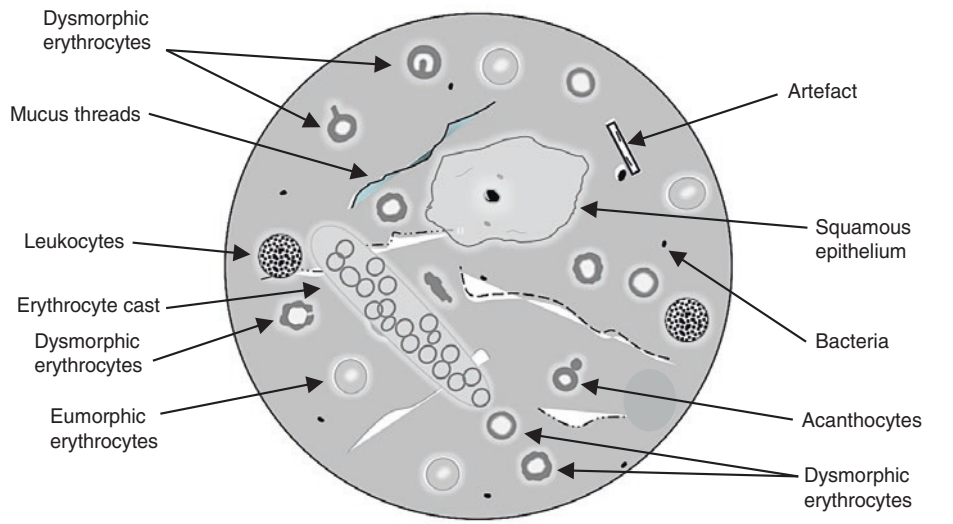


Fig. 12.4 Dysmorphic hematuria → suspected renal hematuria

EumEc: 1-4	/HPF	SqEc: 0-1	/HPF	EumEc: 18	%	100 Ec
DysEc: 5-15	/HPF	Bact: (+)	/HPF	DysEc: 74	%	
Lc: 1-4	/HPF			Acanthocytes: 8	%	

Count the number of eumorphic erythrocytes, dysmorphic erythrocytes, and acanthocytes per 100 erythrocytes and determine the percentage.

12.2.5 Dysmorphic Hematuria with Erythrocyte Casts



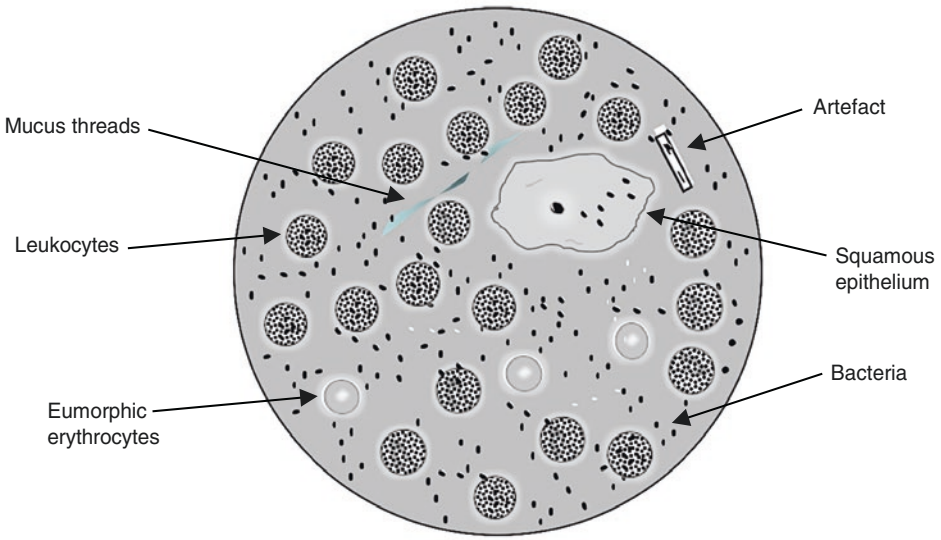
EumEc:	1-4	/HPF	SqEc:	0-1	/HPF	EumEc:	20	%
DysEc:	5-15	/HPF	Bact:	(+)	/HPF	DysEc:	74	%
Lc:	1-4	/HPF	EcCa	2	/aHPF	Acanthocytes:	6	%

100 Ec

Count the number of eumorphic erythrocytes, dysmorphic erythrocytes, and acanthocytes per 100 erythrocytes and determine the percentage.

Fig. 12.5 Dysmorphic hematuria and erythrocyte casts → suspected renal hematuria

12.2.6 Bacterial Urinary Tract Infection



EumEc:	1-4	/HPF	SqEc:	0-1	/HPF
DysEc:		/HPF	Bact:	++	/HPF
Lc:	15-50	/HPF			

Fig. 12.6 Bacterial urinary tract infection

12.2.7 Bacterial Urinary Tract Infections with Renal Involvement

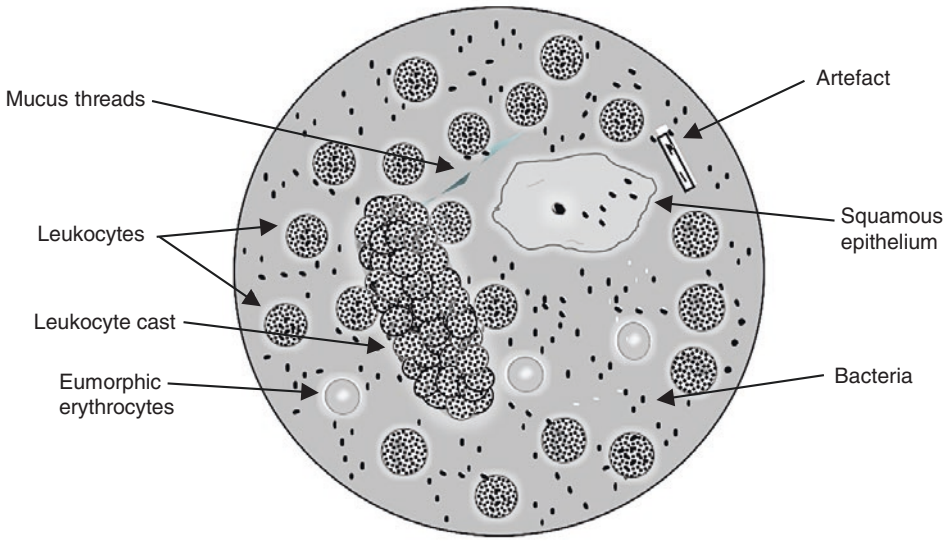


Fig. 12.7 Bacterial urinary tract infection with renal involvement

EumEc:	1-4	/HPF	SqEc:	0-1	/HPF
DysEc:	-	/HPF	Bact:	++	/HPF
Lc:	15-50	/HPF	LcCa:	2	/aHPF

12.2.8 Yeast Infections

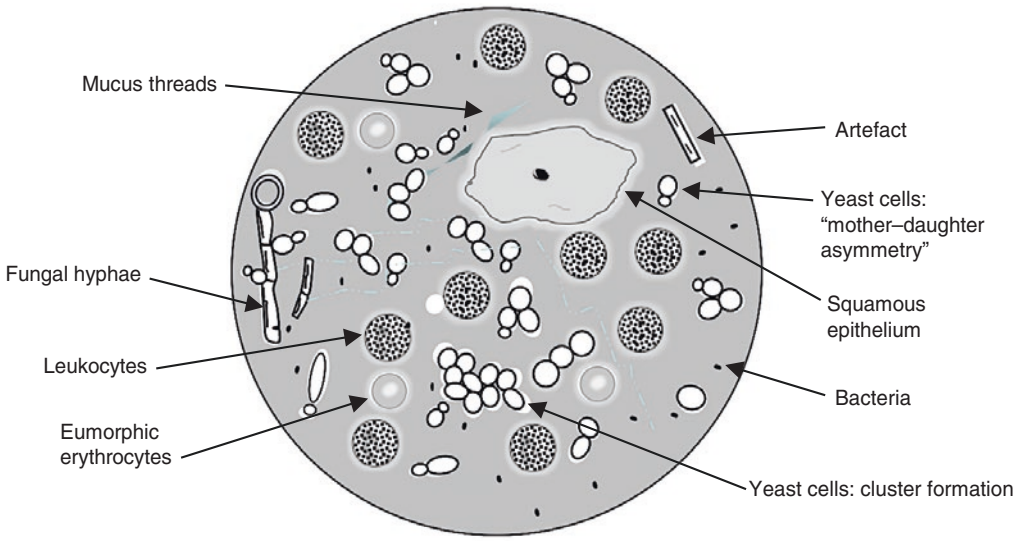


Fig. 12.8 Yeast infection

EumEc:	1-4	/HPF	SqEc:	0-1	/HPF
DysEc:		/HPF	Bact:	(+)	/HPF
Lc:	5-15	/HPF	Yeast cells:	++	/HPF

12.2.9 Yeast Contamination

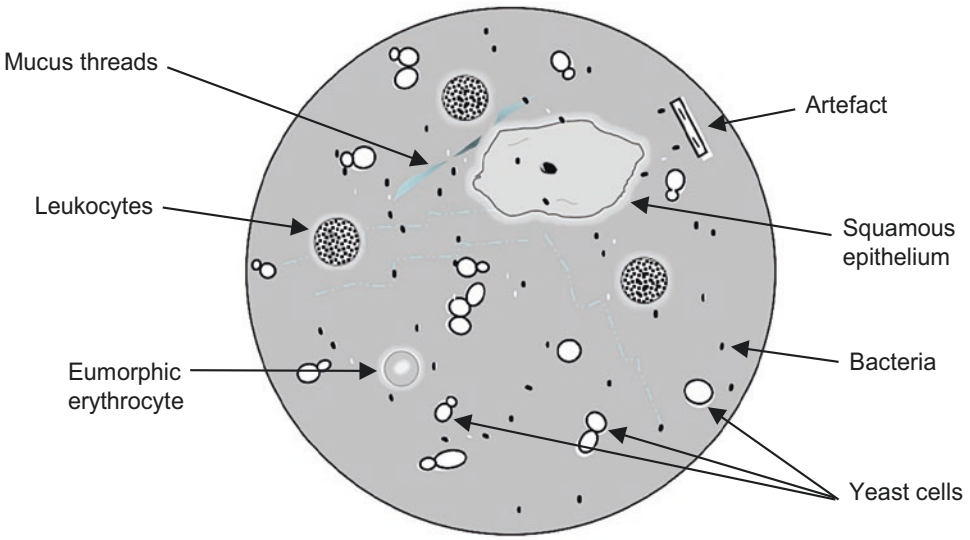


Fig. 12.9 Yeast contamination

EumEc:	0-1	/HPF	SqEc:	0-1	/HPF
DysEc:	-	/HPF	Bact:	(+)-+	/HPF
Lc:	1-4	/HPF	Yeast cells:	(+)	/HPF

12.2.10 Pseudo-urinary Tract Infection

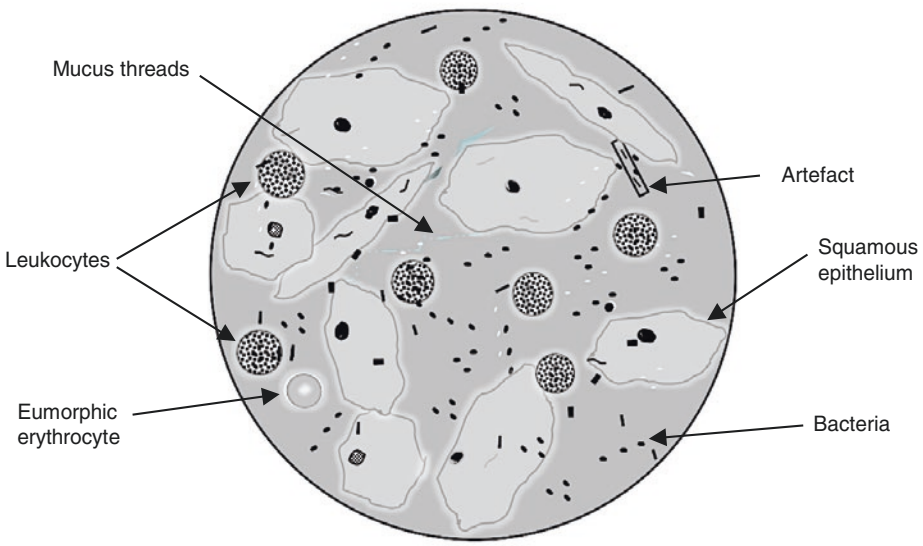


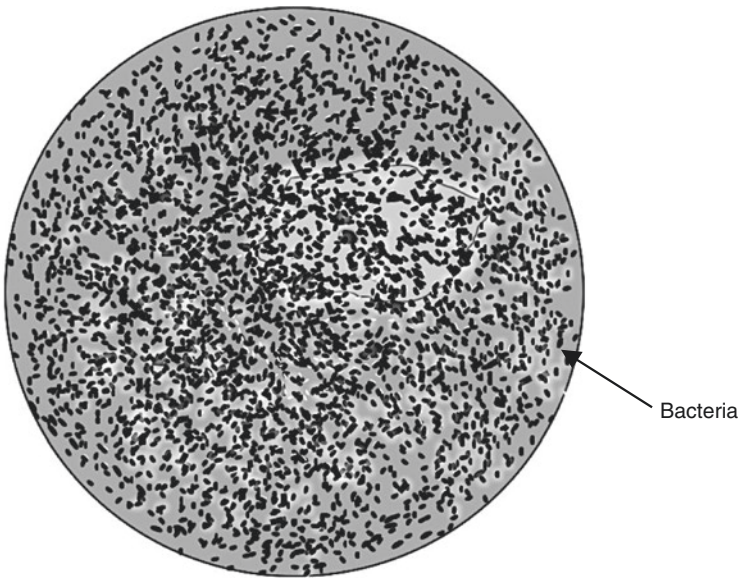
Fig. 12.10 Pseudo-urinary tract infection

EumEc:	0-1	/HPF	SqEc:	5-15	/HPF
DysEc:	-	/HPF	Bact:	+-++	/HPF
Lc:	5-15	/HPF			

Explanation: If more than seven to eight squamous epithelial cells are counted per HPF (18 mm field number), one can assume that the urine sample was not taken from midstream urine. Therefore, it is likely that the increased leukocytes, bacteria, and squamous epithelial cells originate from the outer genital tract and not the urinary tract.

12.2.11 Bacteriuria

Fig. 12.11 Bacteriuria



EumEc:	0-1	/HPF	SqEc:	0-1	/HPF
DysEc:	-	/HPF	Bact:	+++	/HPF
Lc:	0-1	/HPF			

12.3 Analysis

12.3.1 Exercises in the microscopic analysis of urinary sediment images

For didactic and technical reasons, the photographs shown below are of different sizes. Despite the different HPF sizes of the individual images, the analysis of urine constituents is given in the "(/HPF)" unit. All images were taken at the same microscopic resolution (400× magnification/18 mm field number).

12.3.2 Eumorphic Hematuria

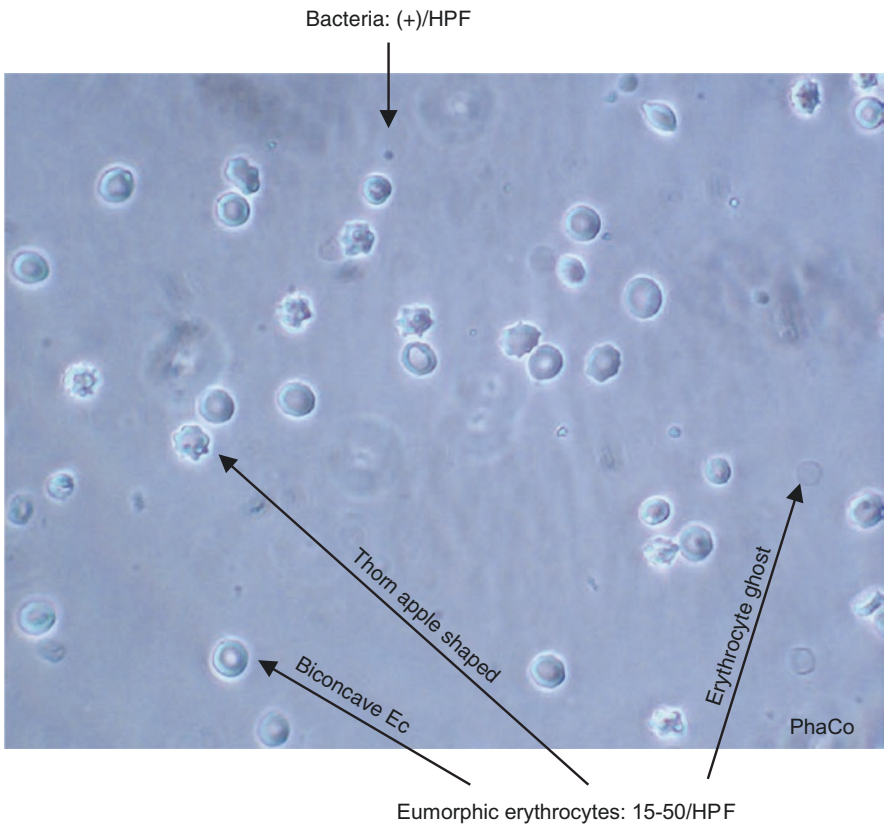


Fig. 12.12 Eumorphic hematuria

12.3.3 Eumorphic Hematuria and Yeast Cells

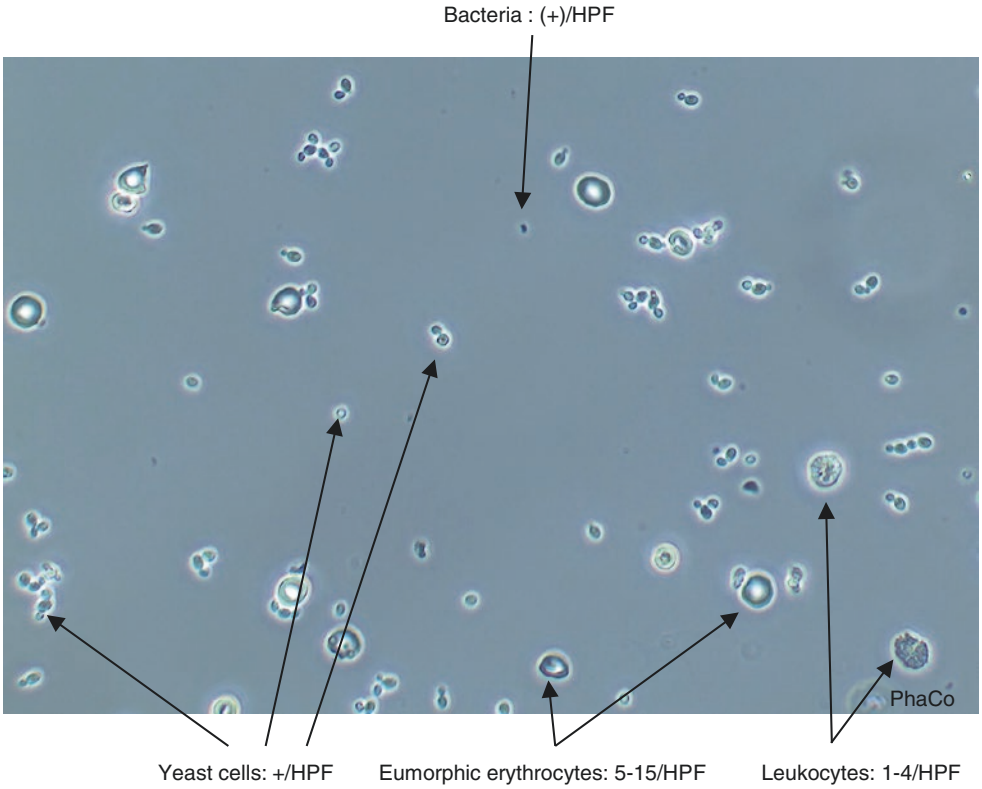


Fig. 12.13 Eumorphic hematuria and yeast cells

12.3.4 Eumorphic Hematuria and Yeast Cells with Fungal Hyphae

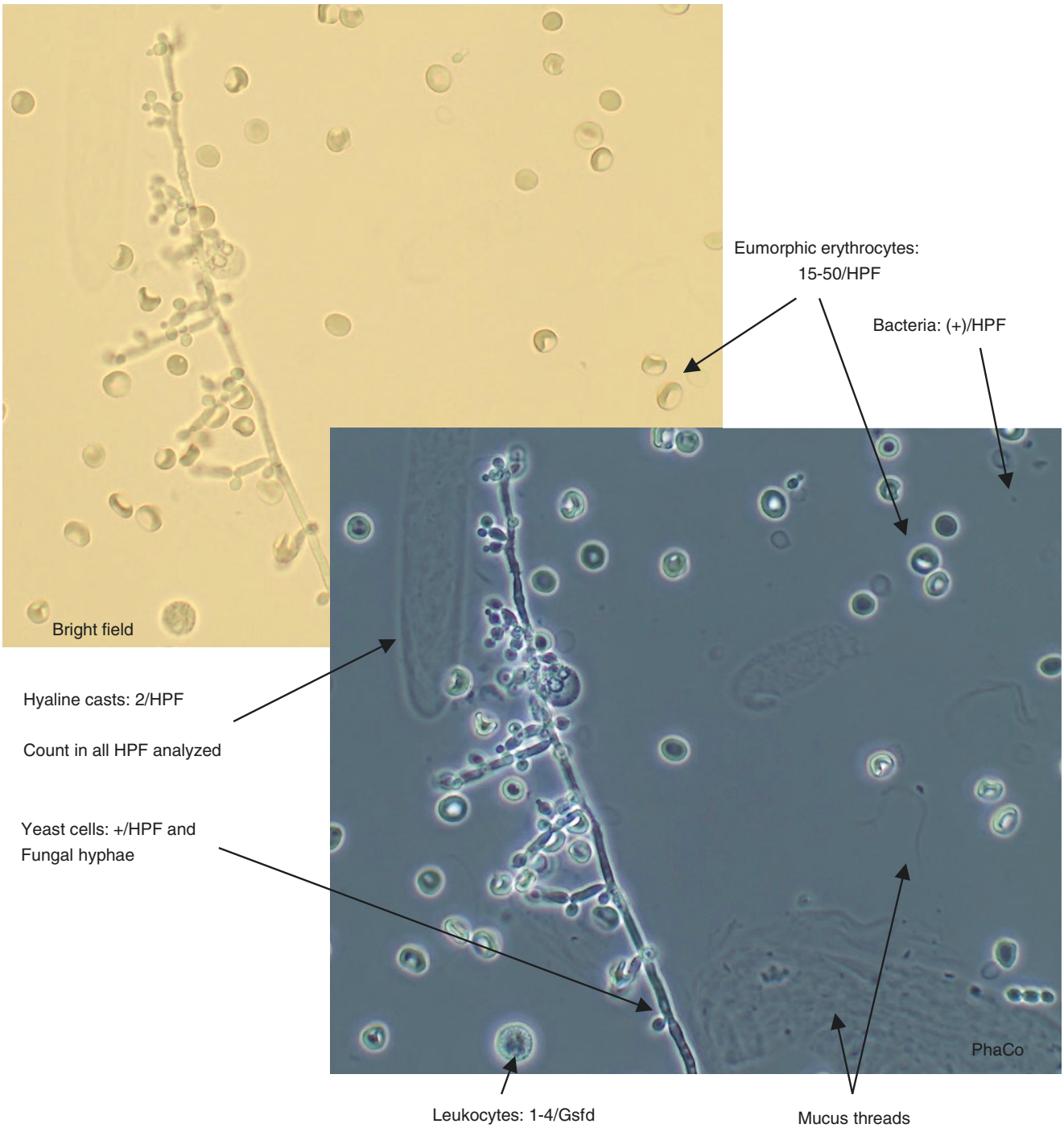


Fig. 12.14 Eumorphic hematuria and yeast cells with fungal hyphae

12.3.5 Eumorphic Hematuria with Crystalluria

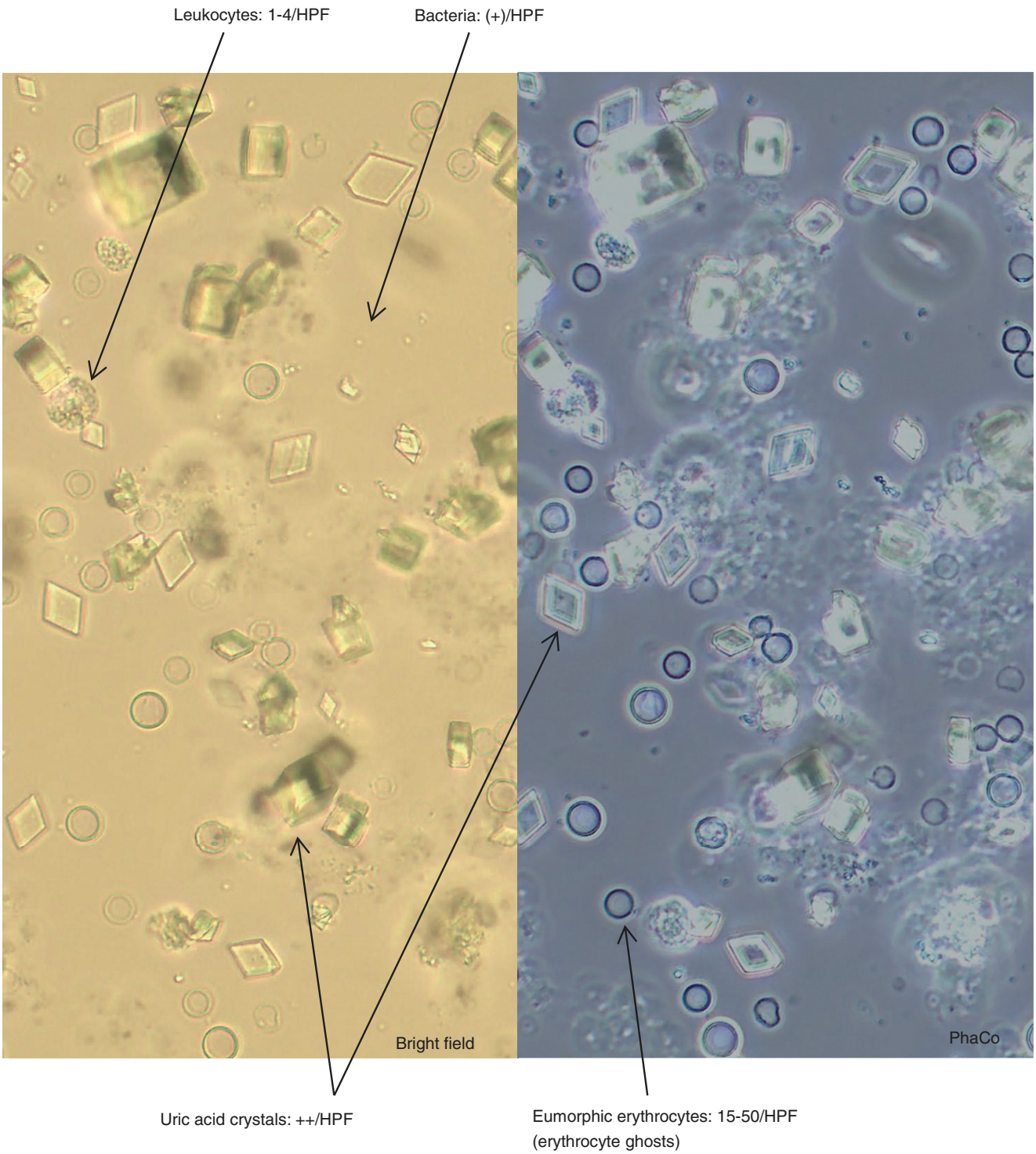


Fig. 12.15 Eumorphic hematuria with crystalluria (uric acid crystals)

12.3.6 Dysmorphic Hematuria

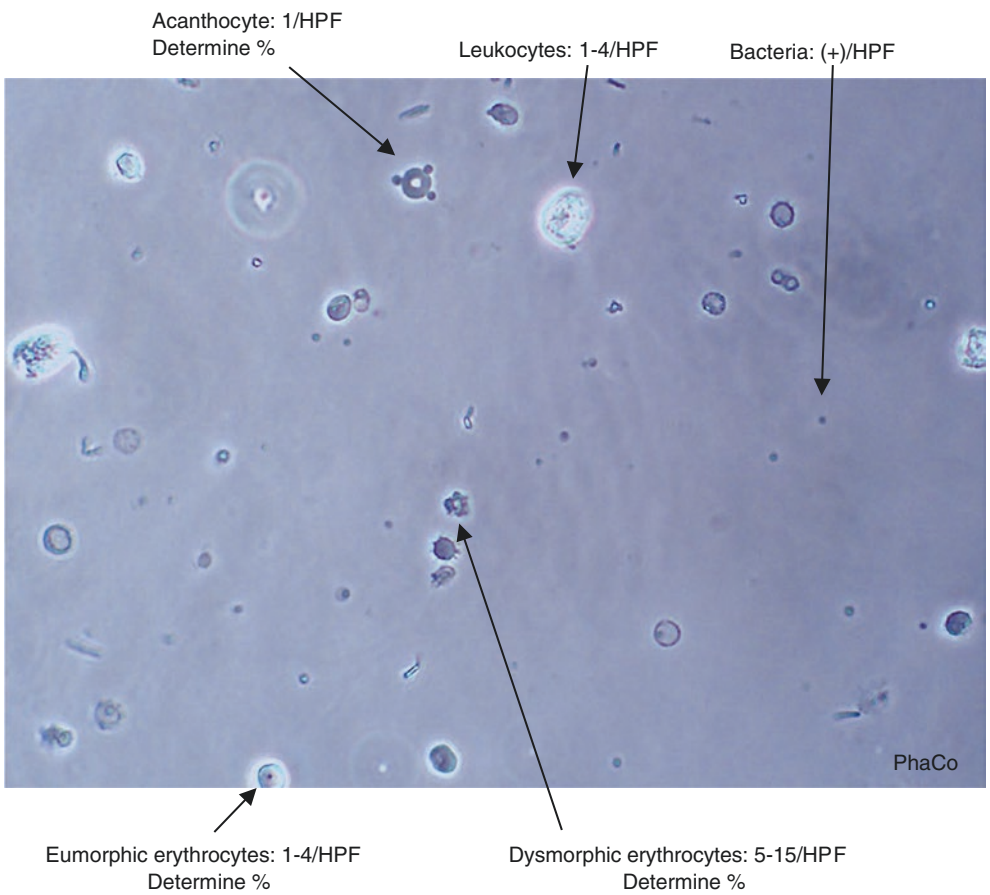
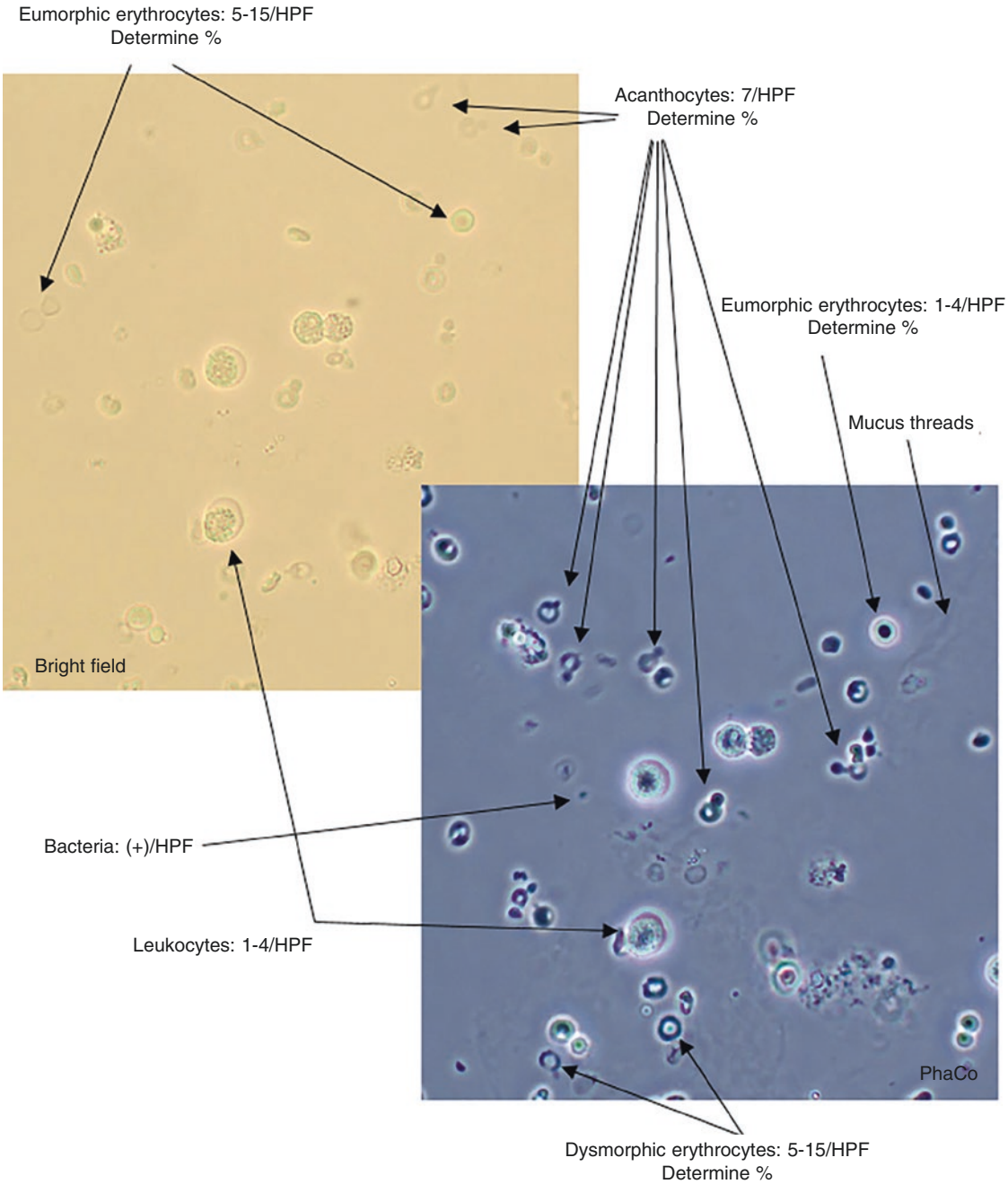


Fig. 12.16 Dysmorphic hematuria I

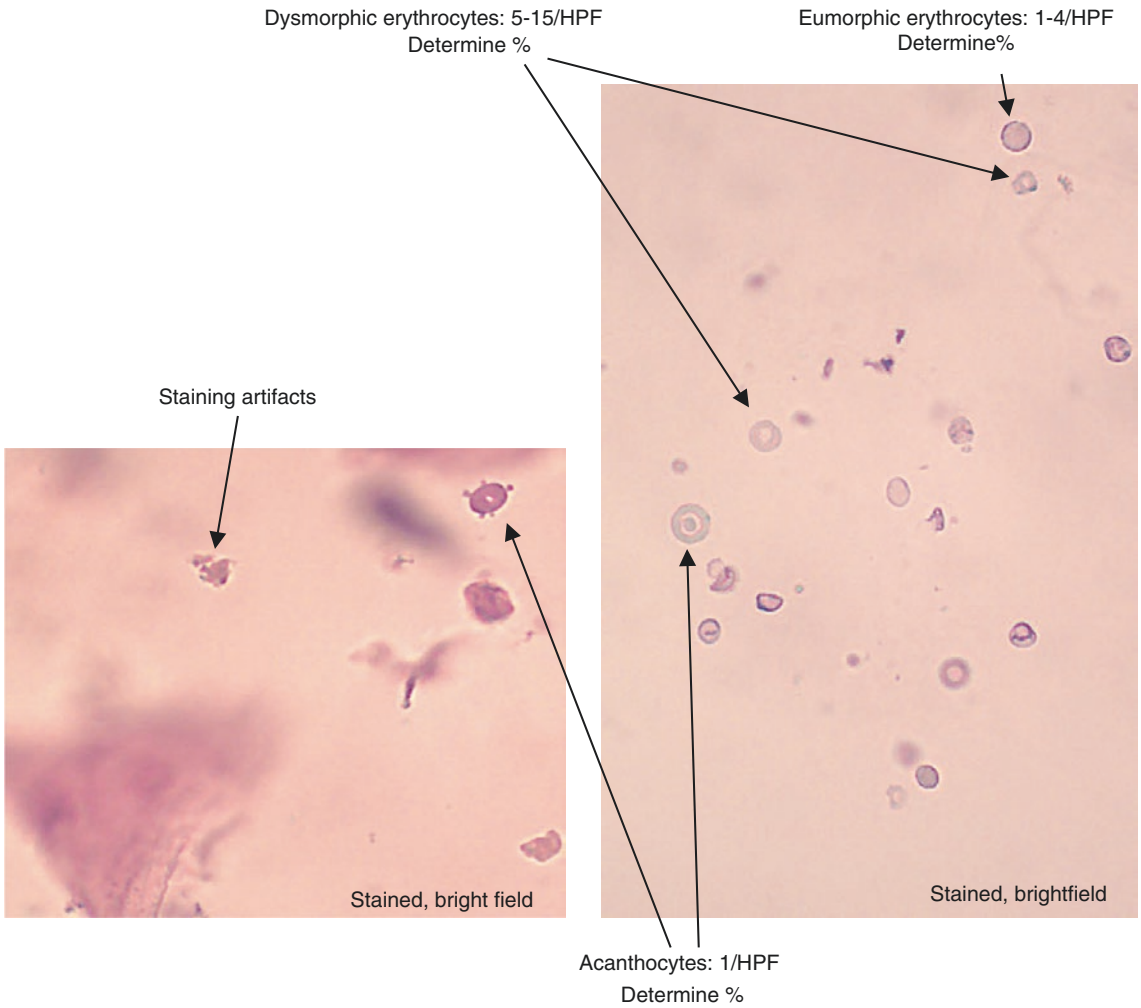
Count the number of eumorphic erythrocytes, dysmorphic erythrocytes, and acanthocyte per 100 erythrocytes and determine the percentage!



Count the number of eumorphic erythrocytes, dysmorphic erythrocytes, and acanthocytes per 100 erythrocytes and determine the percentage!

Fig. 12.17 Dysmorphic hematuria II

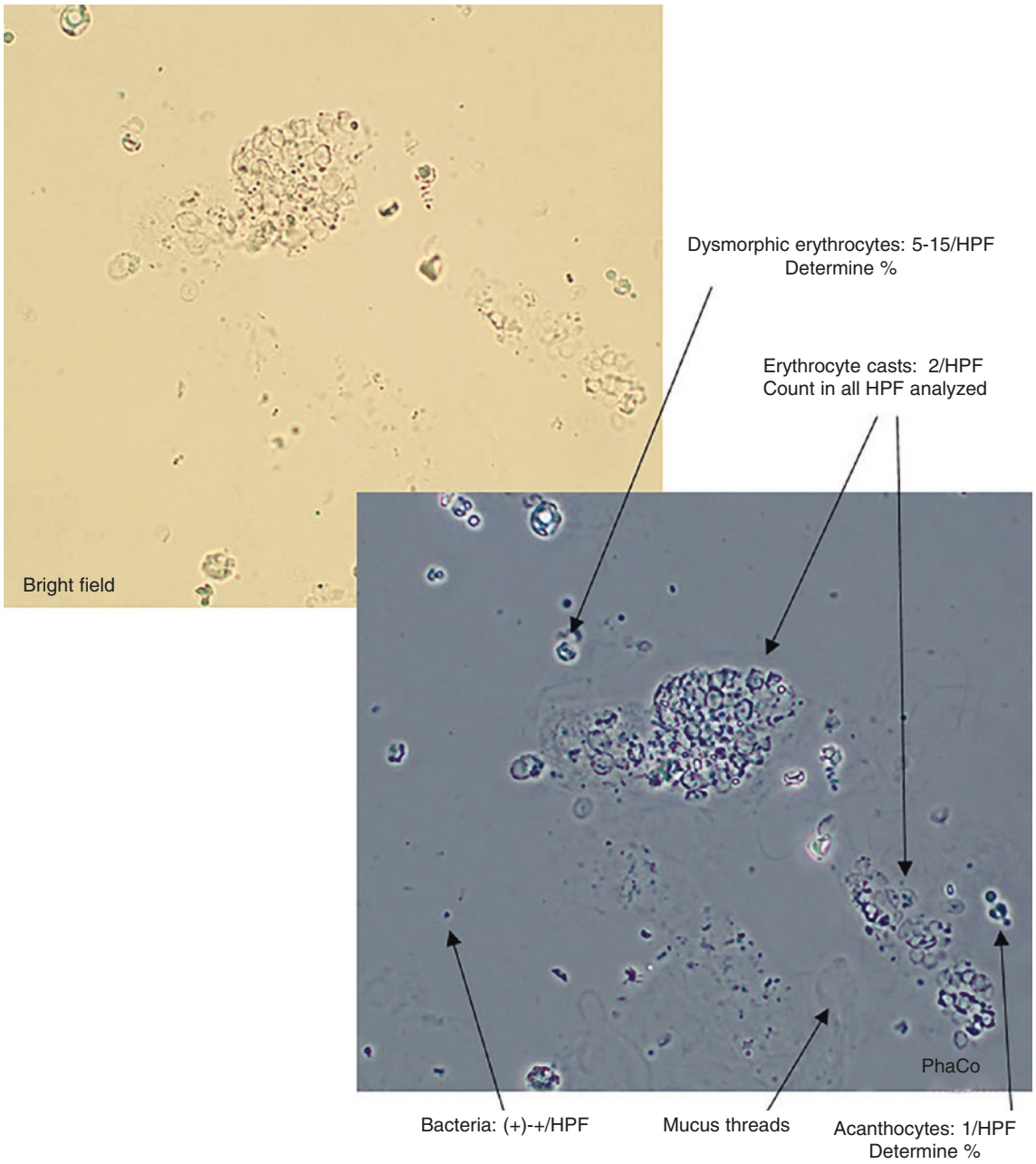
12.3.7 Dysmorphic Hematuria: Stained



Count the number of eumorphic erythrocytes, dysmorphic erythrocytes, and acanthocytes per 100 erythrocytes and determine the percentage!

Fig. 12.18 Dysmorphic hematuria III stained with KOVA® stain reagent

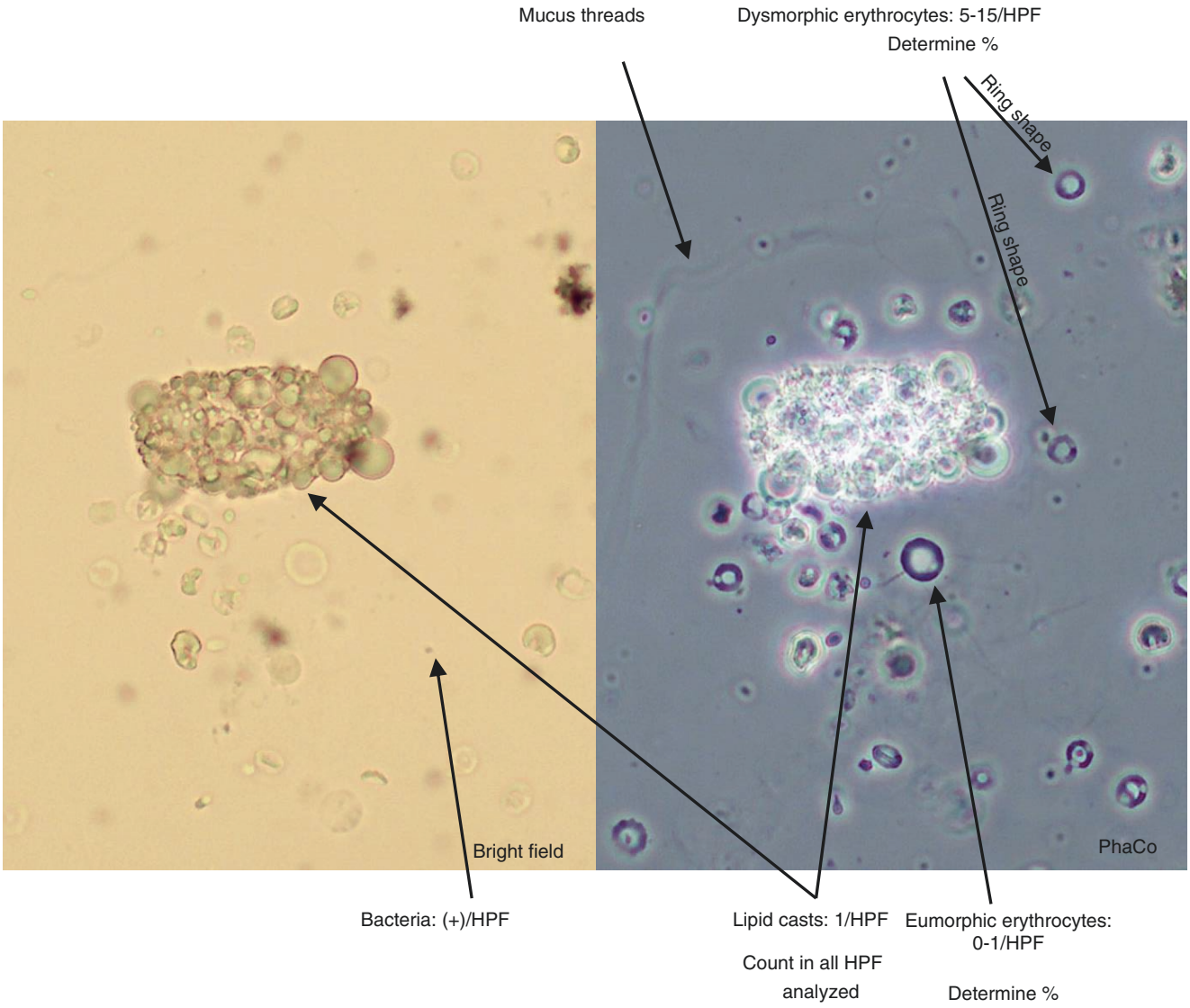
12.3.8 Dysmorphic Hematuria and Erythrocyte Casts



Count the number of eumorphic erythrocytes, dysmorphic erythrocytes, and acanthocytes per 100 erythrocytes and determine the percentage!

Fig. 12.19 Dysmorphic hematuria and erythrocyte casts

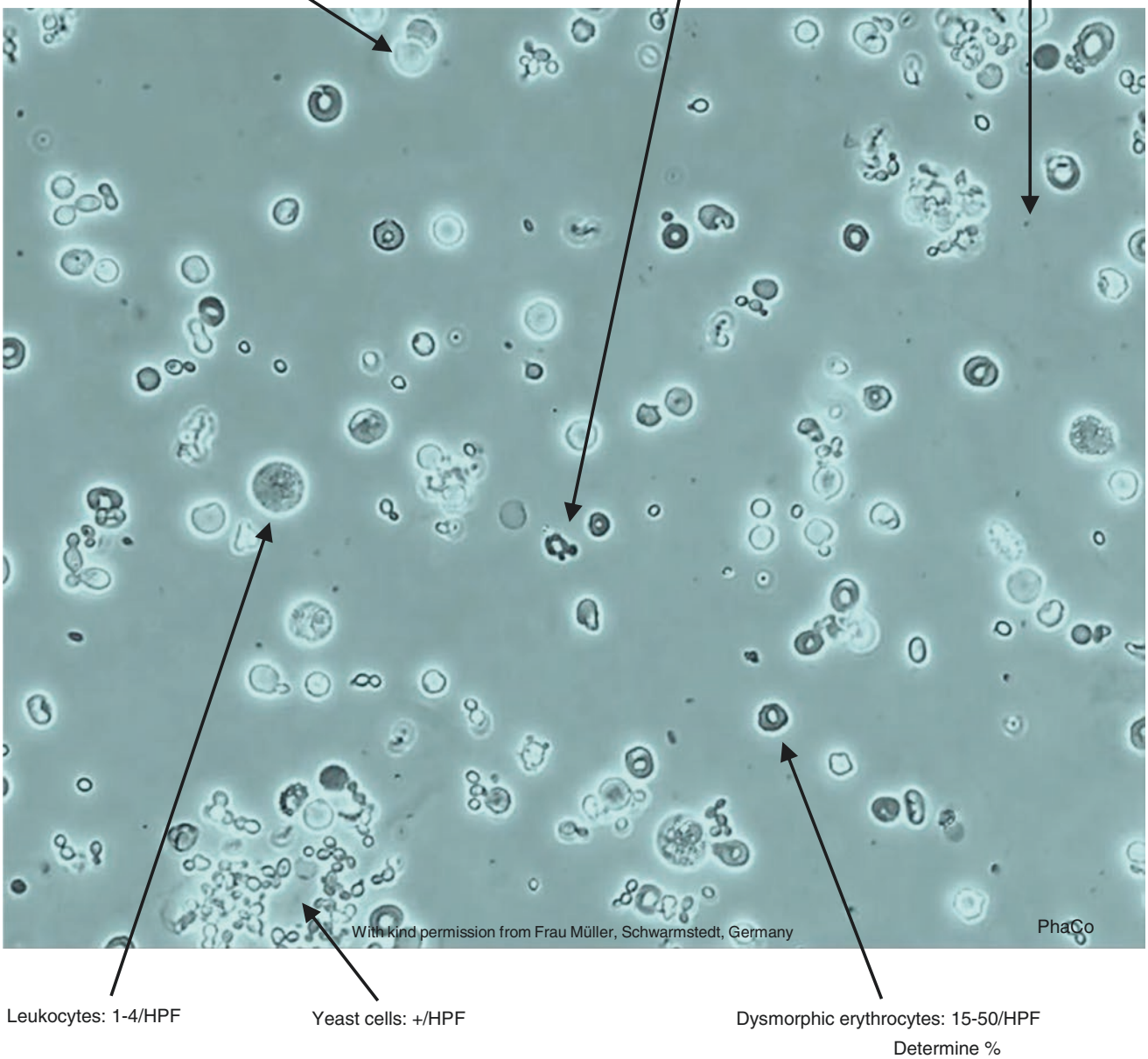
12.3.9 Dysmorphic Hematuria and Lipid Casts



Count the number of eumorphic erythrocytes, dysmorphic erythrocytes, and acanthocytes per 100 erythrocytes and determine the percentage!

Fig. 12.20 Dysmorphic hematuria and lipid casts

12.3.10 Dysmorphic Hematuria with Yeast Cells



Count the number of eumorphic erythrocytes, dysmorphic erythrocytes, and acanthocytes per 100 erythrocytes and determine the percentage!

Fig. 12.21 Dysmorphic hematuria with yeast cells

12.3.11 Leukocyturia

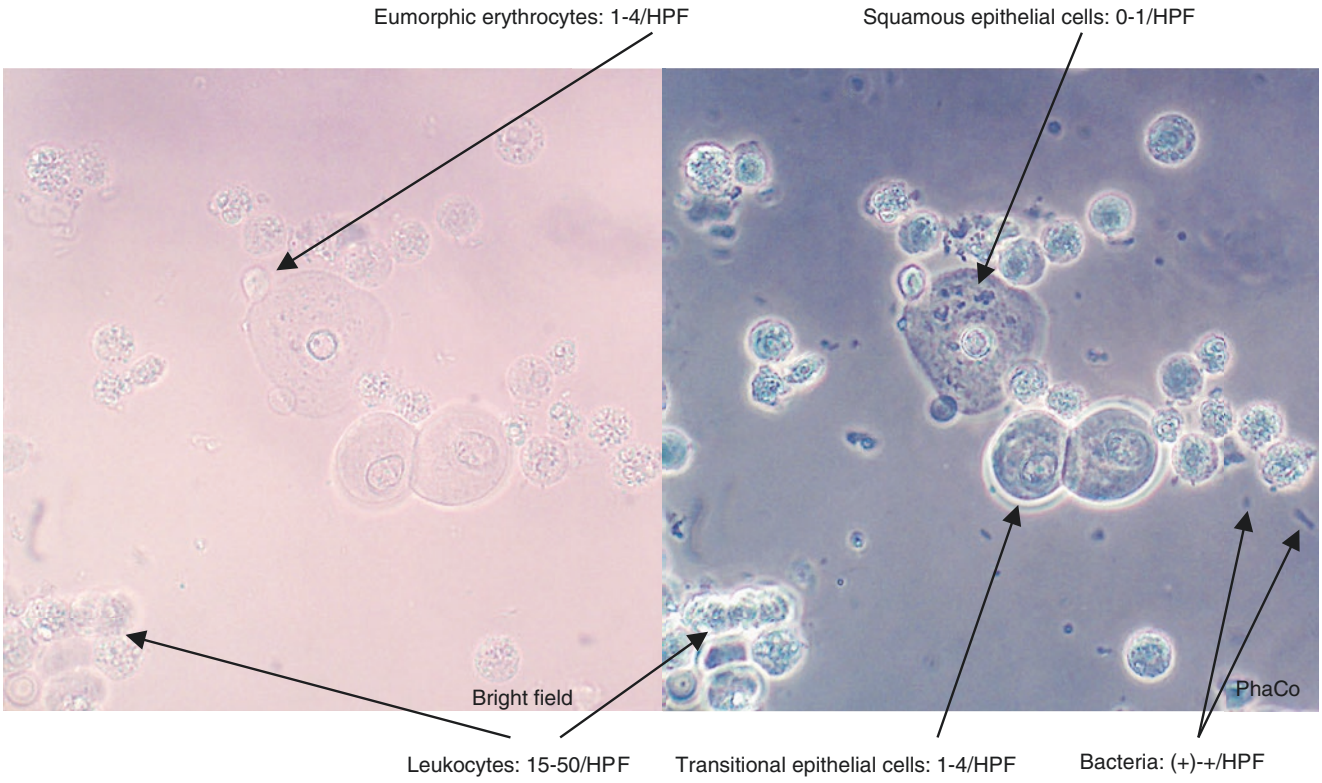


Fig. 12.22 Leukocyturia I

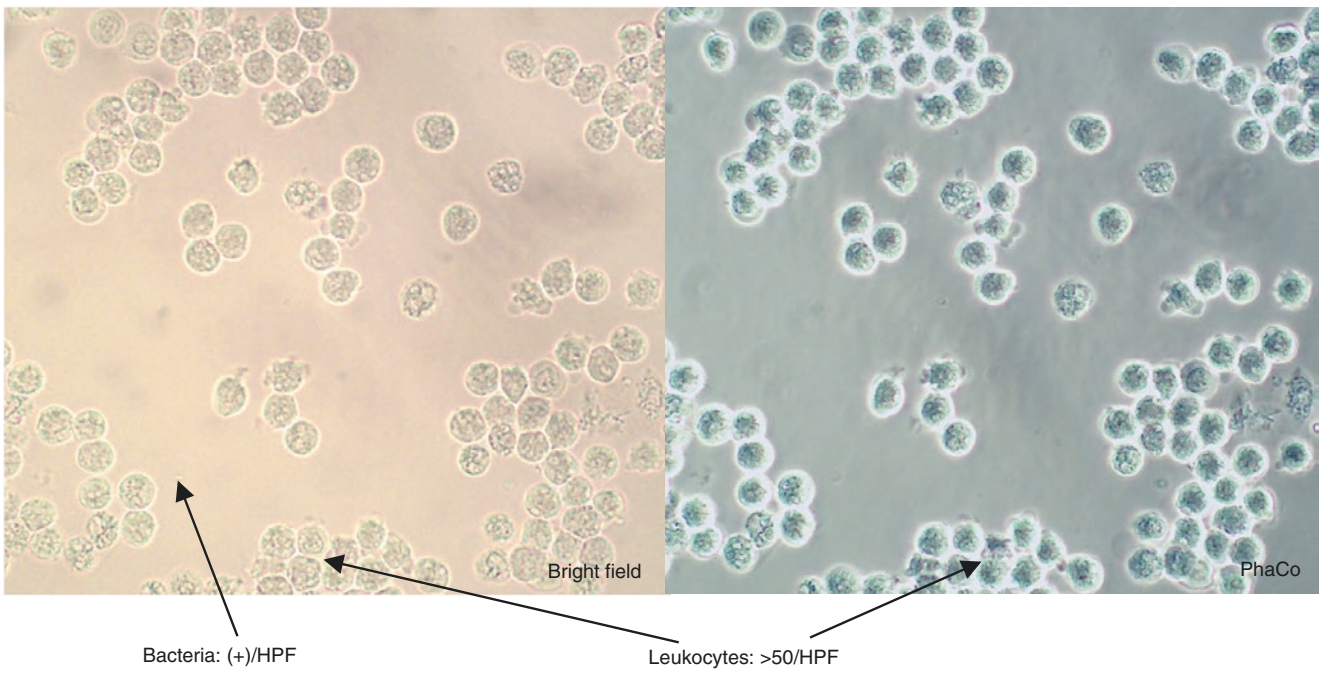


Fig. 12.23 Leukocyturia II

12.3.12 Leukocyturia and Bacteriuria

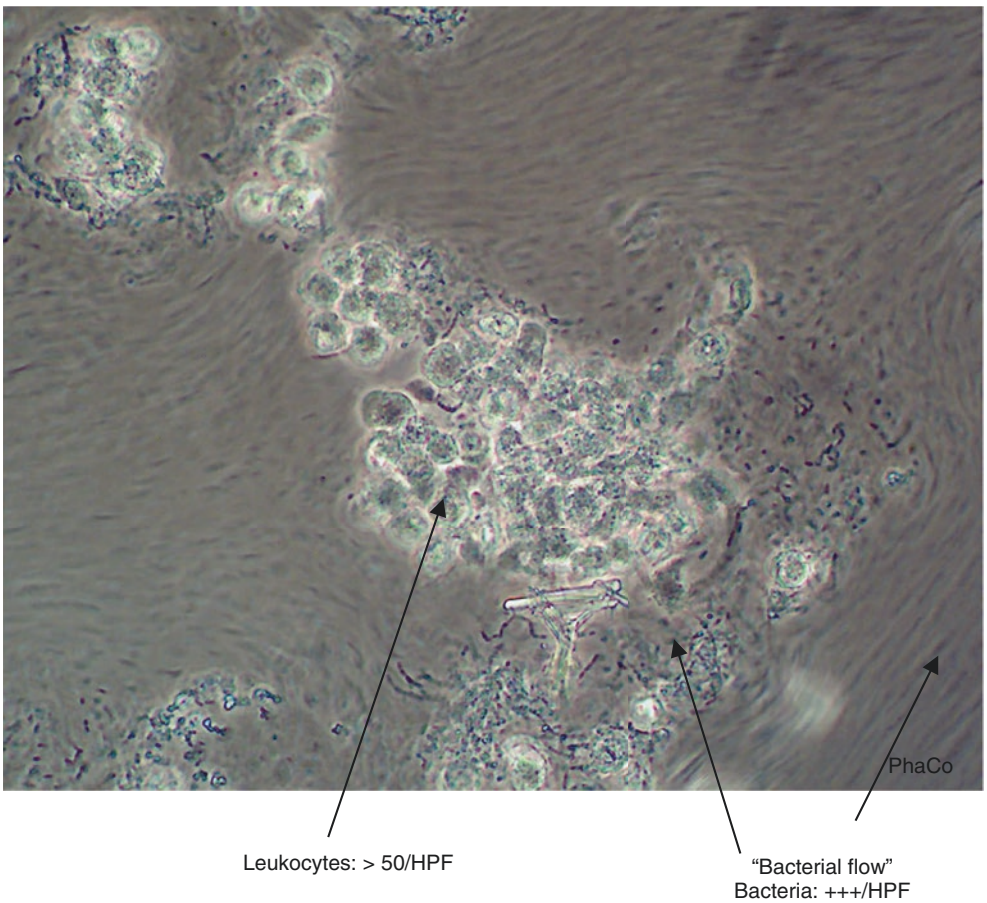


Fig. 12.24 Leukocyturia and bacteriuria

12.3.13 Leukocyturia, Bacteriuria, and Triple Phosphates

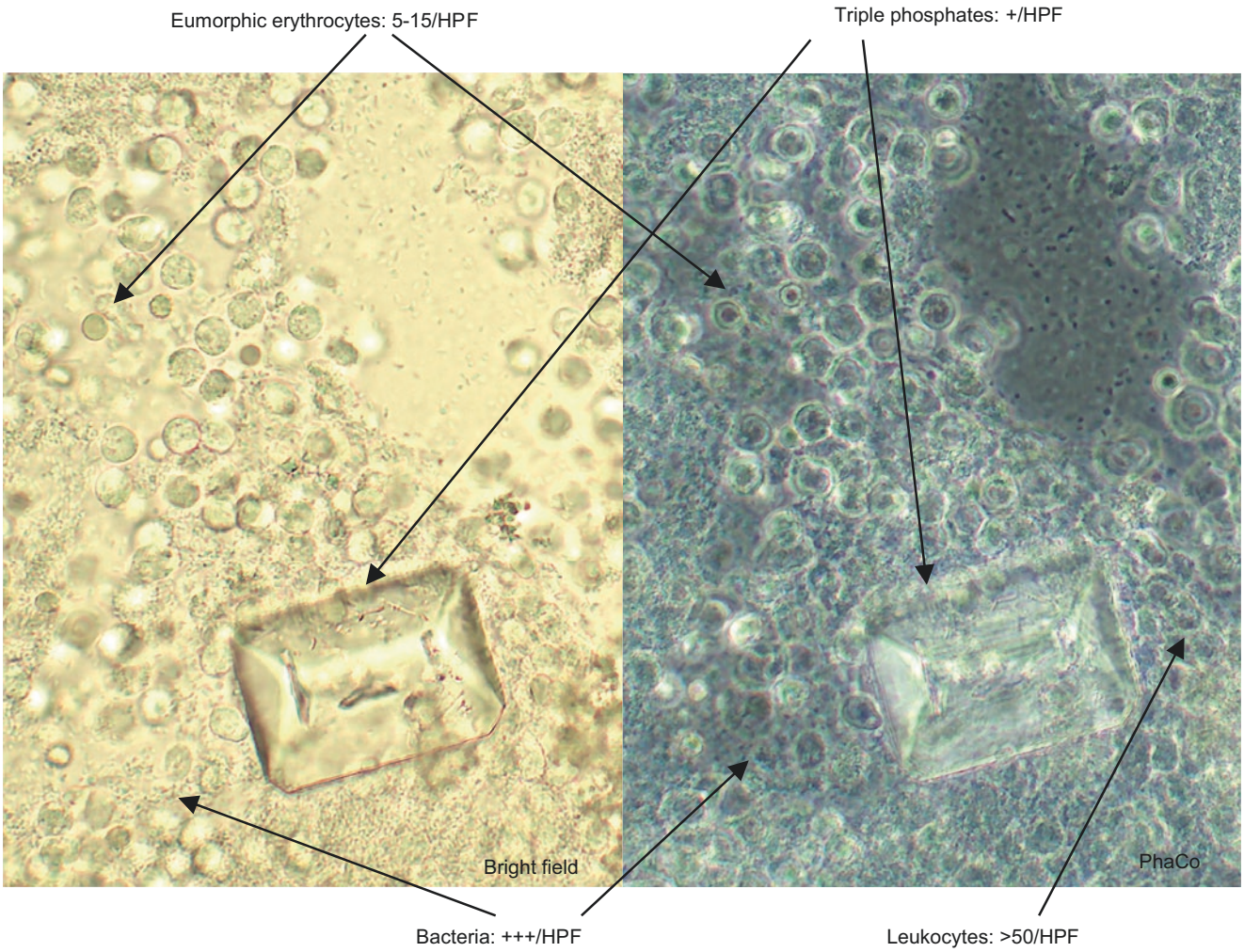


Fig. 12.25 Leukocyturia, bacteriuria, triple phosphates and eumorphic hematuria

12.3.14 Leukocyturia with Leukocyte Casts

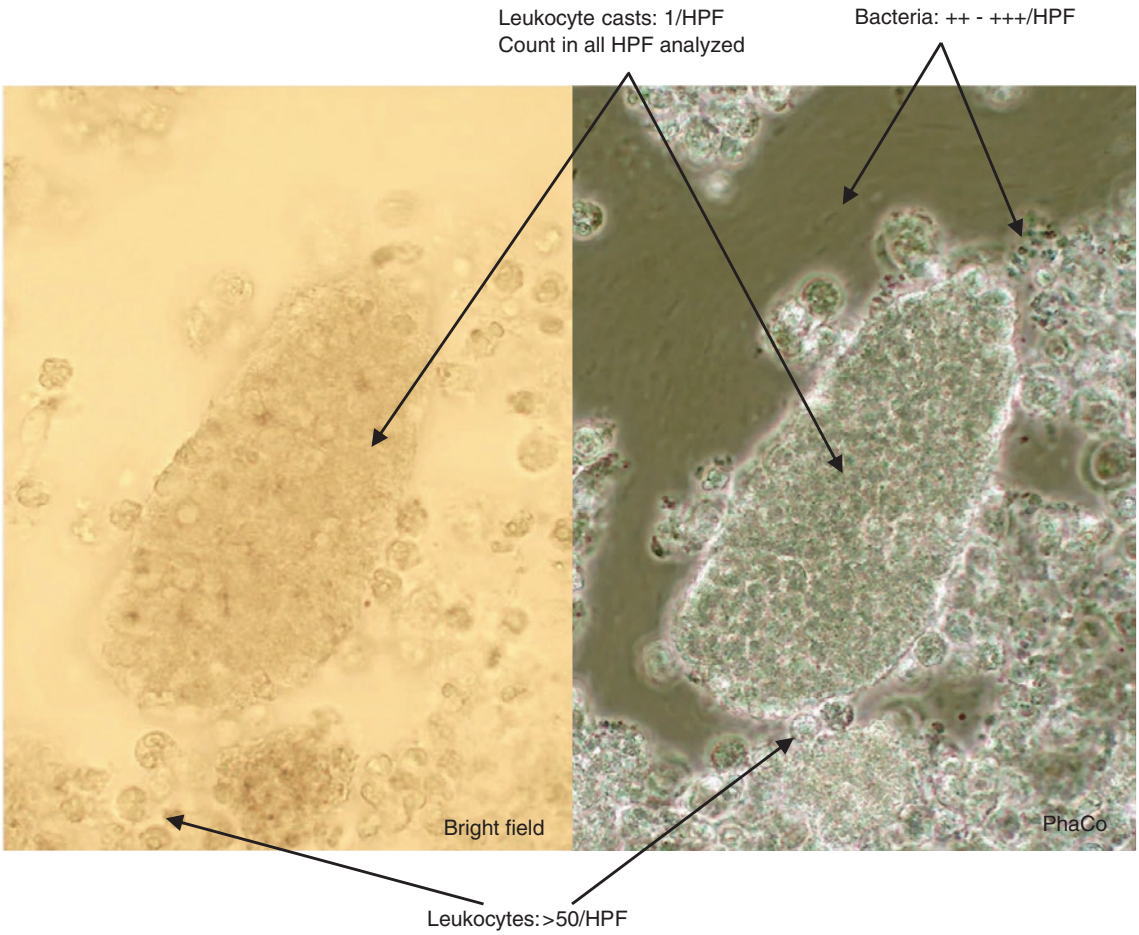


Fig. 12.26 Leukocyturia with leukocyte casts

12.3.15 Leukocyturia and Yeasts

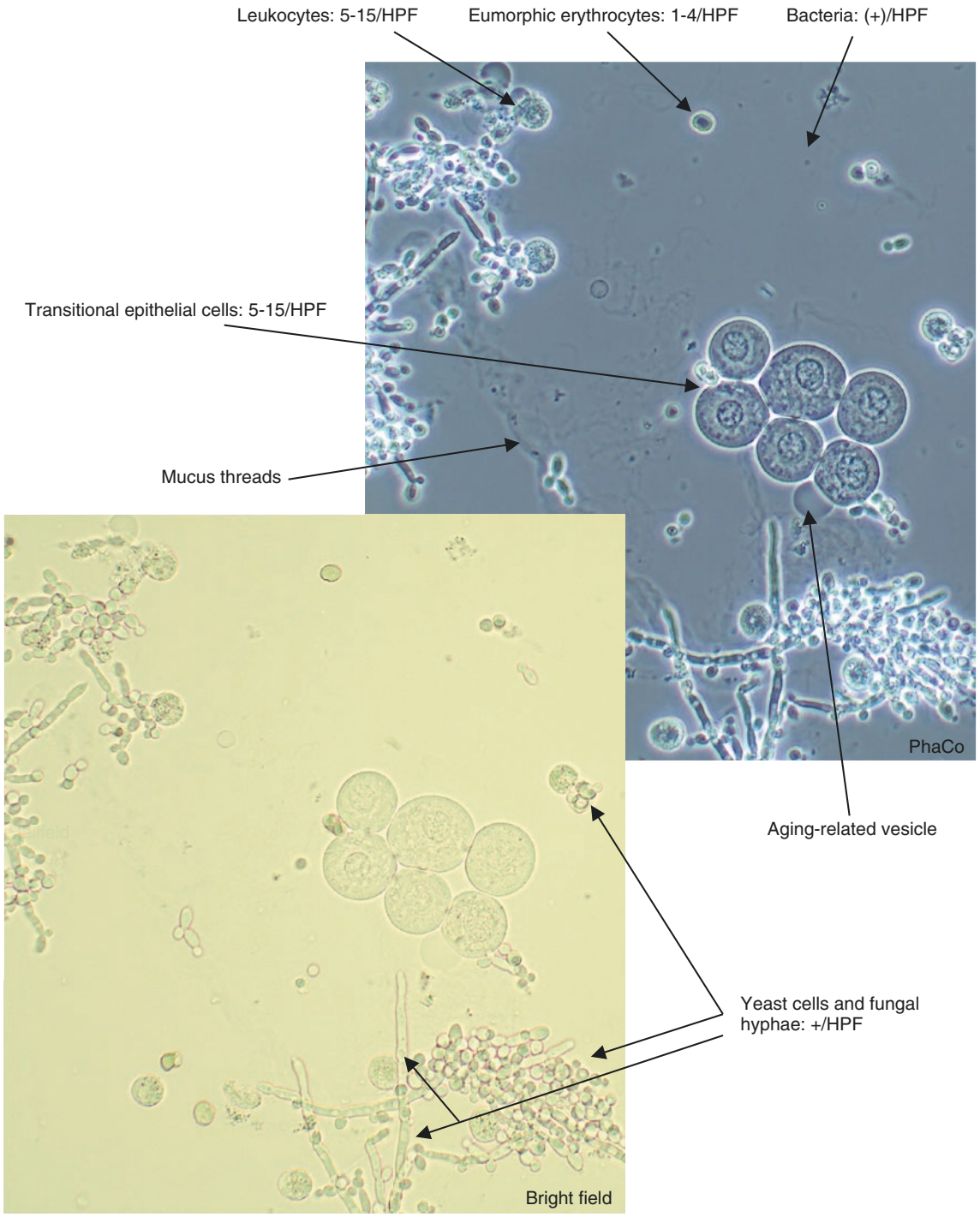


Fig. 12.27 Leukocyturia, yeasts and eumorphic hematuria

12.3.16 Leukocyturia and Spermatozoa

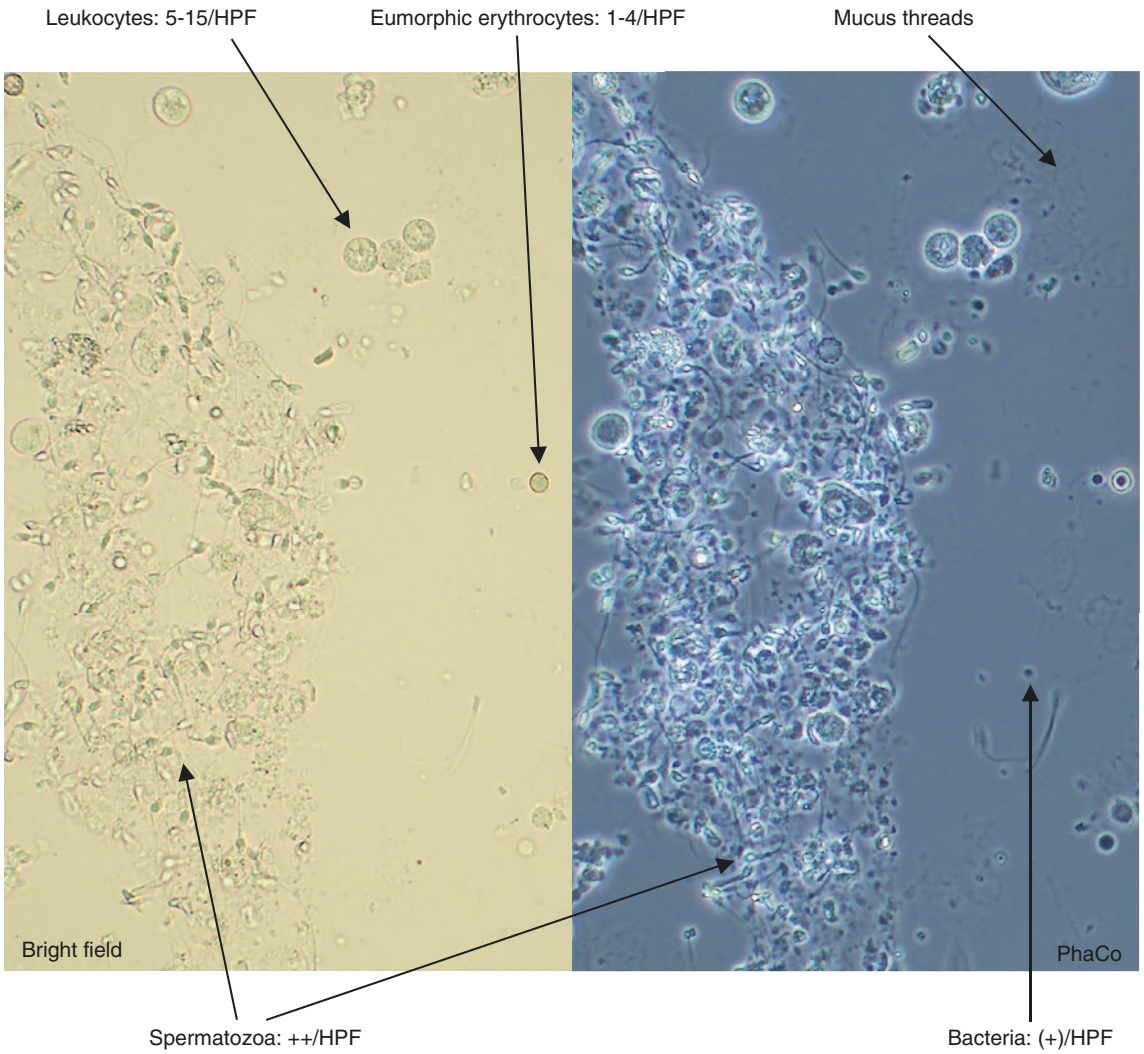


Fig. 12.28 Leukocyturia, spermatozoa and eumorphic hematuria

12.3.17 Bacteriuria and Crystalluria

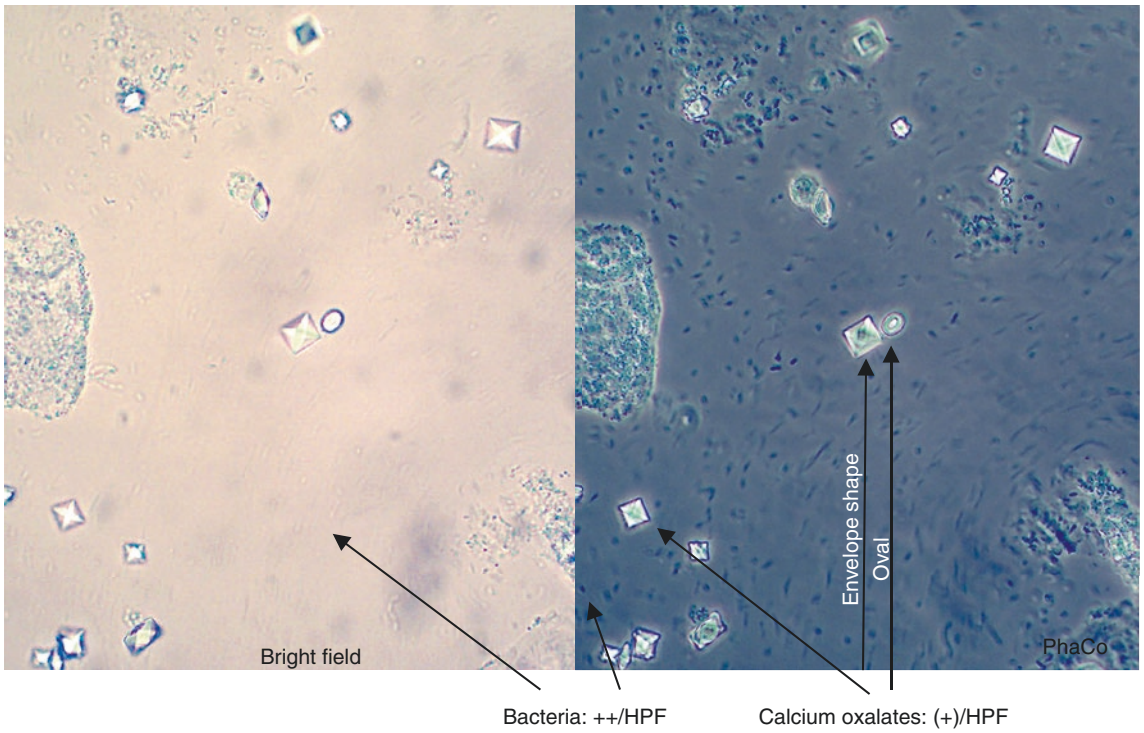


Fig. 12.29 Bacteriuria and crystalluria I (calcium oxalates)

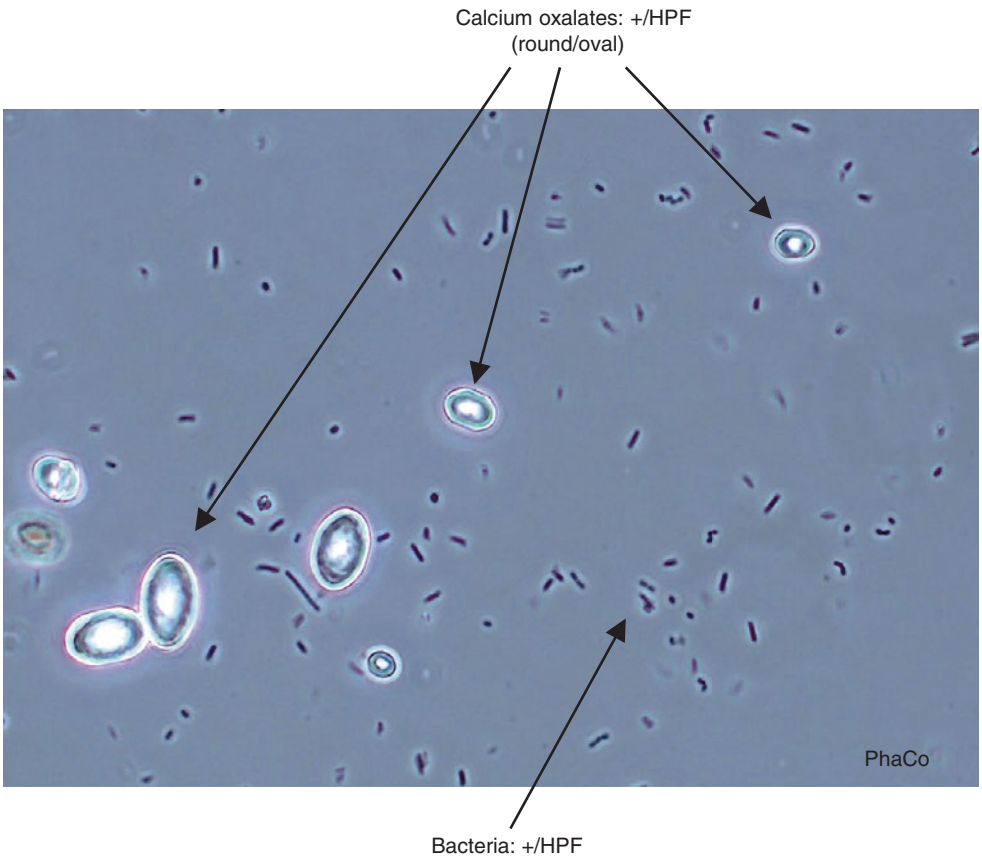


Fig. 12.30 Bacteriuria and crystalluria II (calcium oxalates)

Tip: Round/oval calcium oxalates must not be confused with eumorphic erythrocytes. If fine-focusing with the micrometer knob is constantly operated, calcium oxalates light up brightly, in contrast to the erythrocytes.

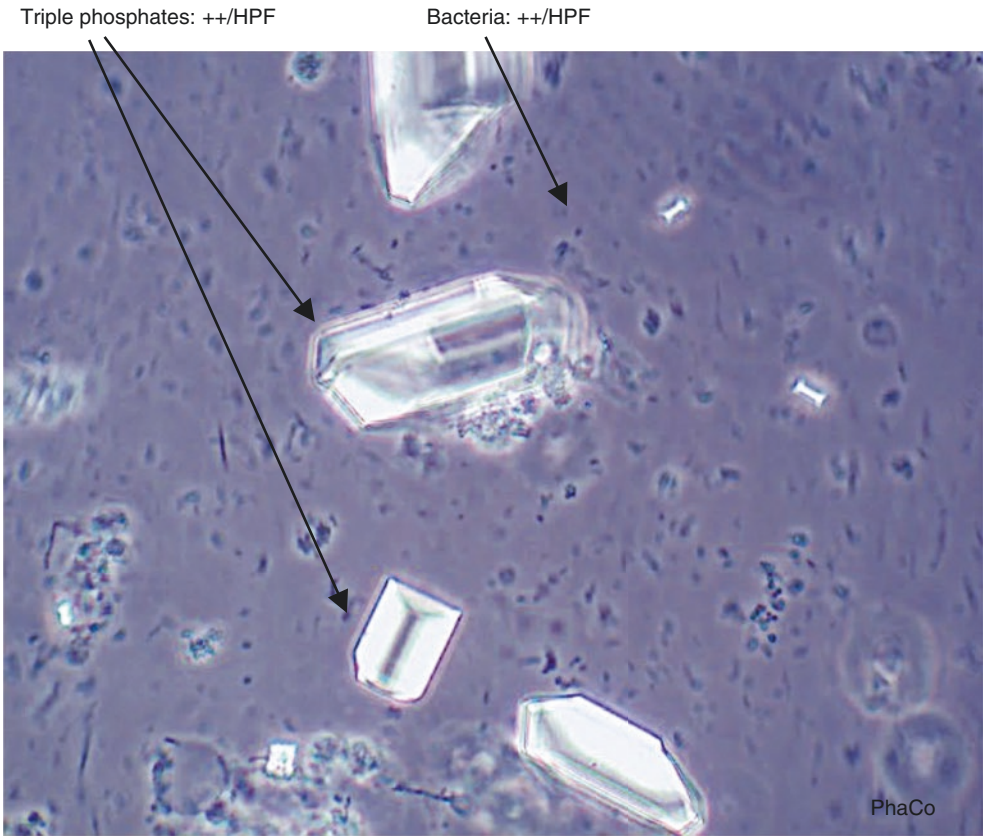


Fig. 12.31 Bacteriuria and crystalluria III (triple phosphates)

Fig. 12.32 Bacteriuria and cristalluria IV (triple phosphates and ammonium urates)

12.3.18 Bacteriuria and Lipiduria

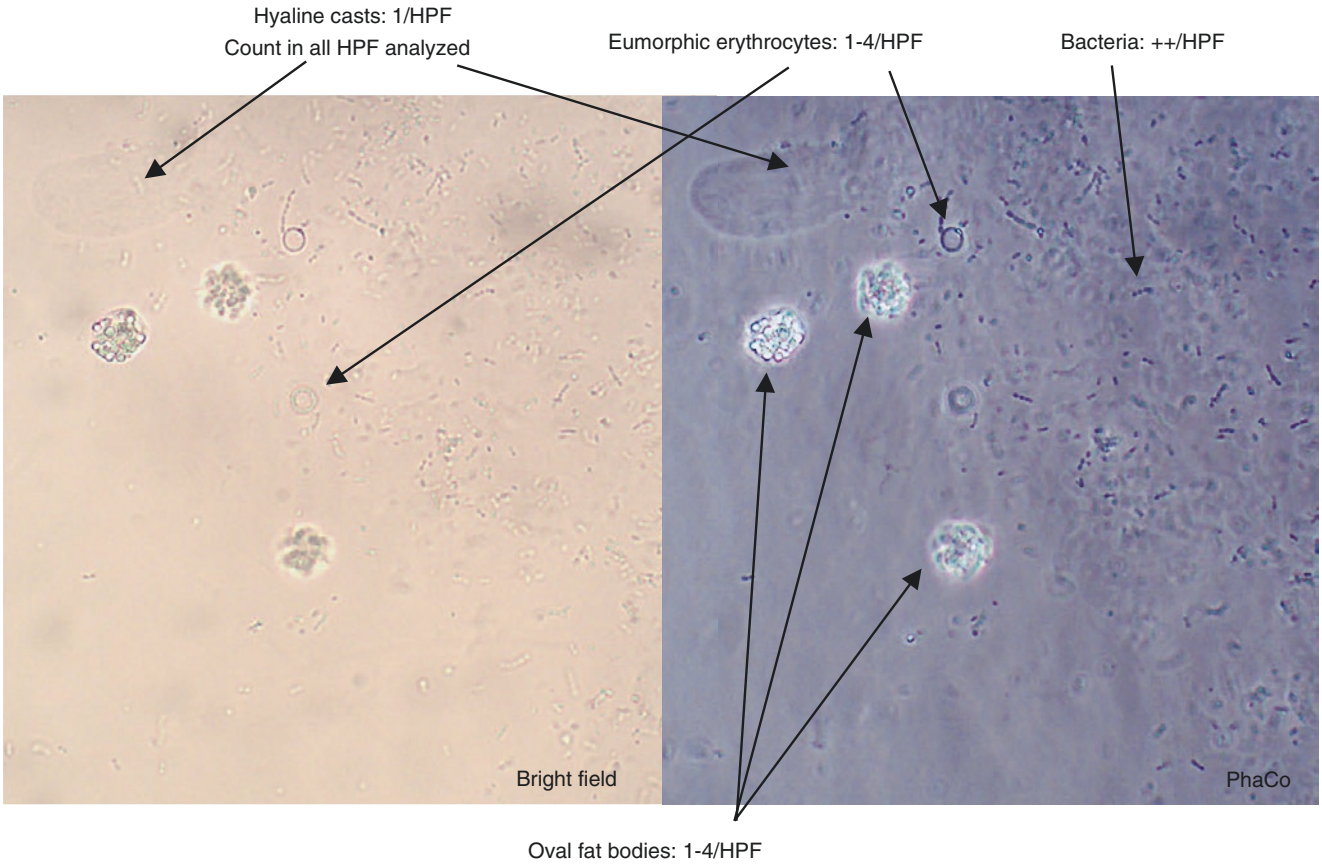
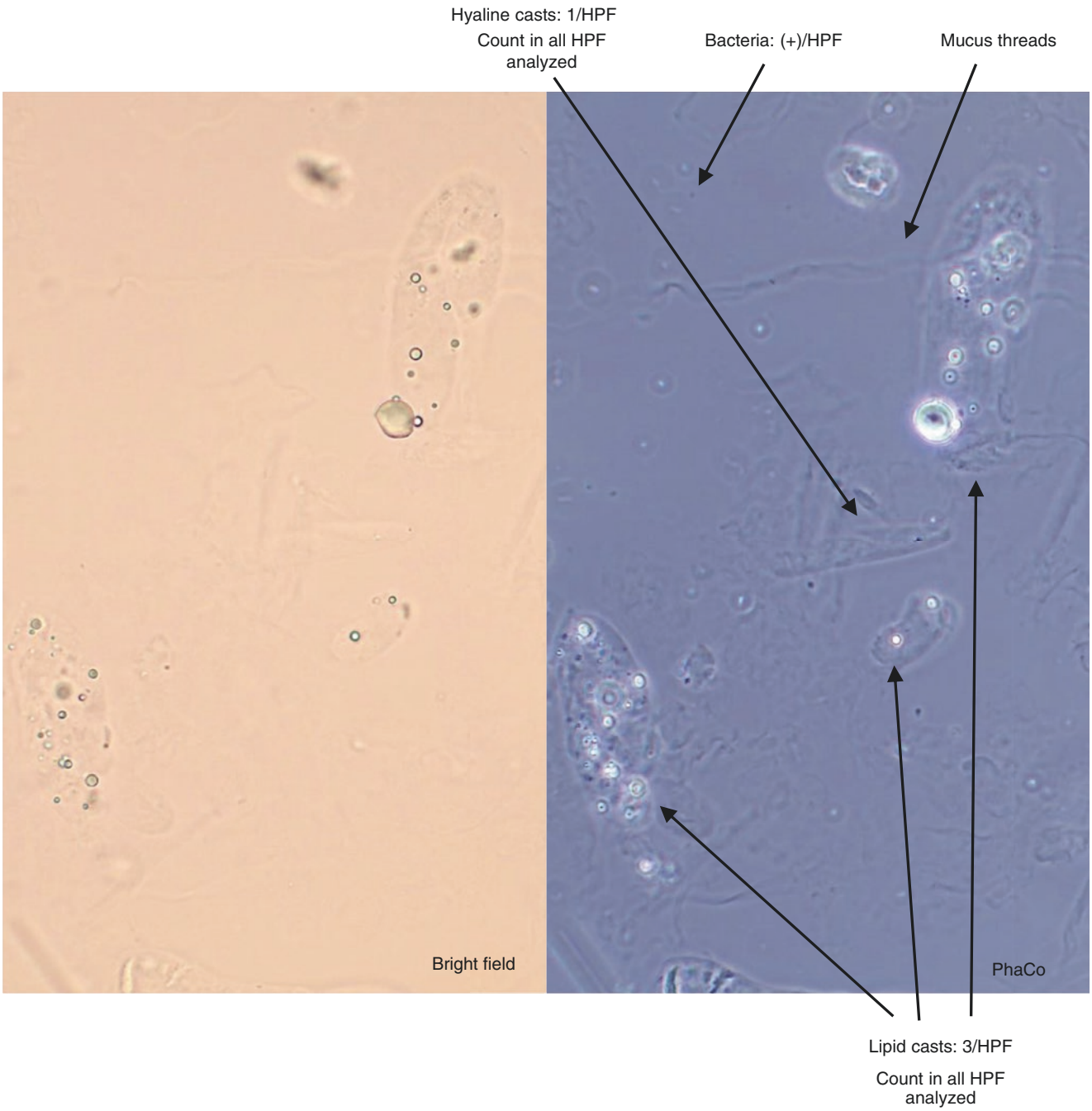


Fig. 12.33 Bacteriuria and lipiduria

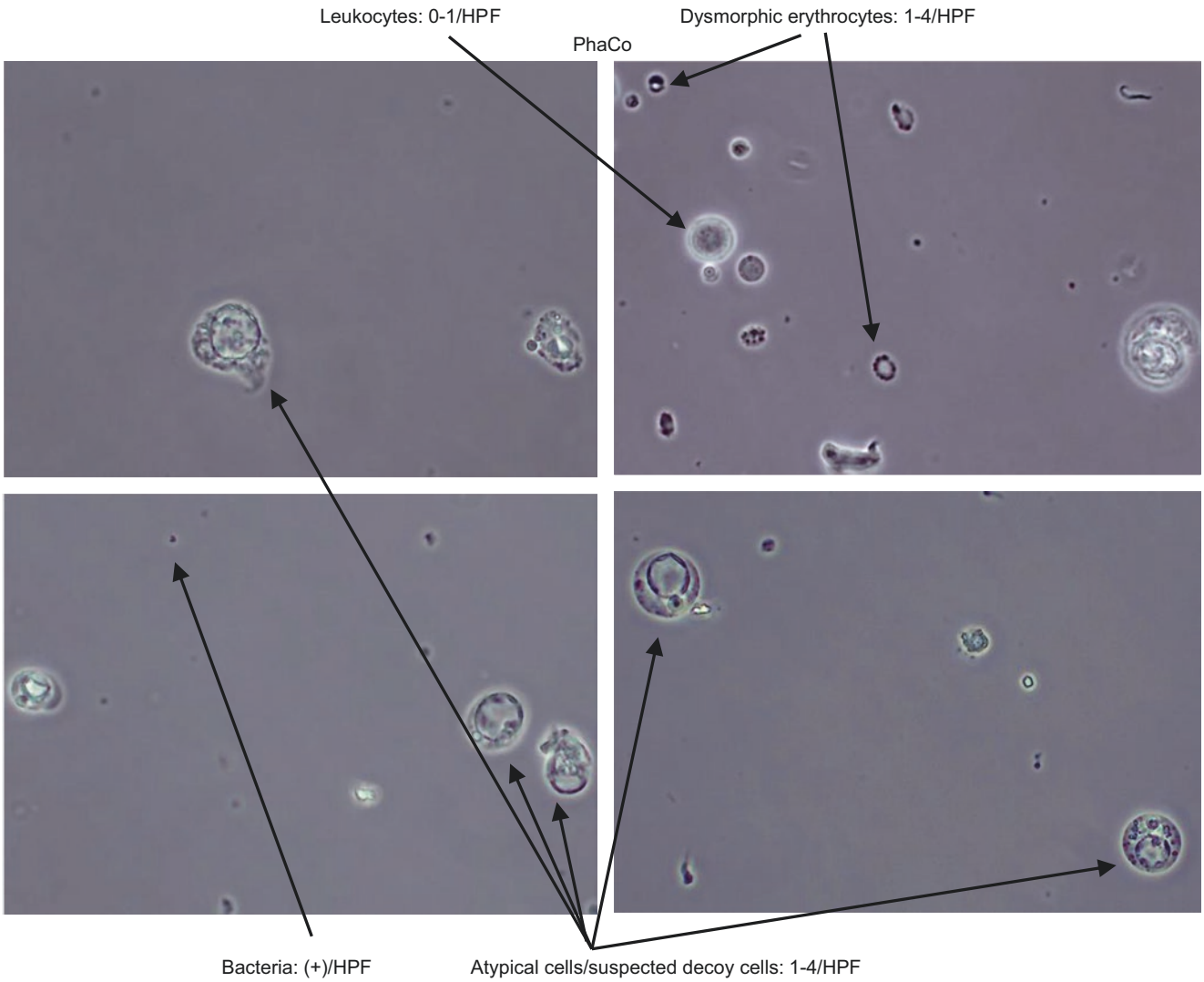
12.3.19 Lipid Cylinduria



Tip: By constantly operating fine-focusing with the micrometer knob, lipid droplets light up strongly and can thus be distinguished from erythrocytes or erythrocyte casts.

Fig. 12.34 Lipid cylinduria

12.3.20 Atypical Cells: Suspected Decoy Cells



Count the number of eumorphic erythrocytes, dysmorphic erythrocytes, and acanthocytes per 100 erythrocytes and determine the percentage!

Tip: One can express only a suspicion of decoy cells, since atypical-looking epithelial cells could also be tumor cells or obsolete cells/epithelial cells.

Fig. 12.35 Suspected decoy cells

12.3.21 Crystalluria and Lipid Casts: Stained

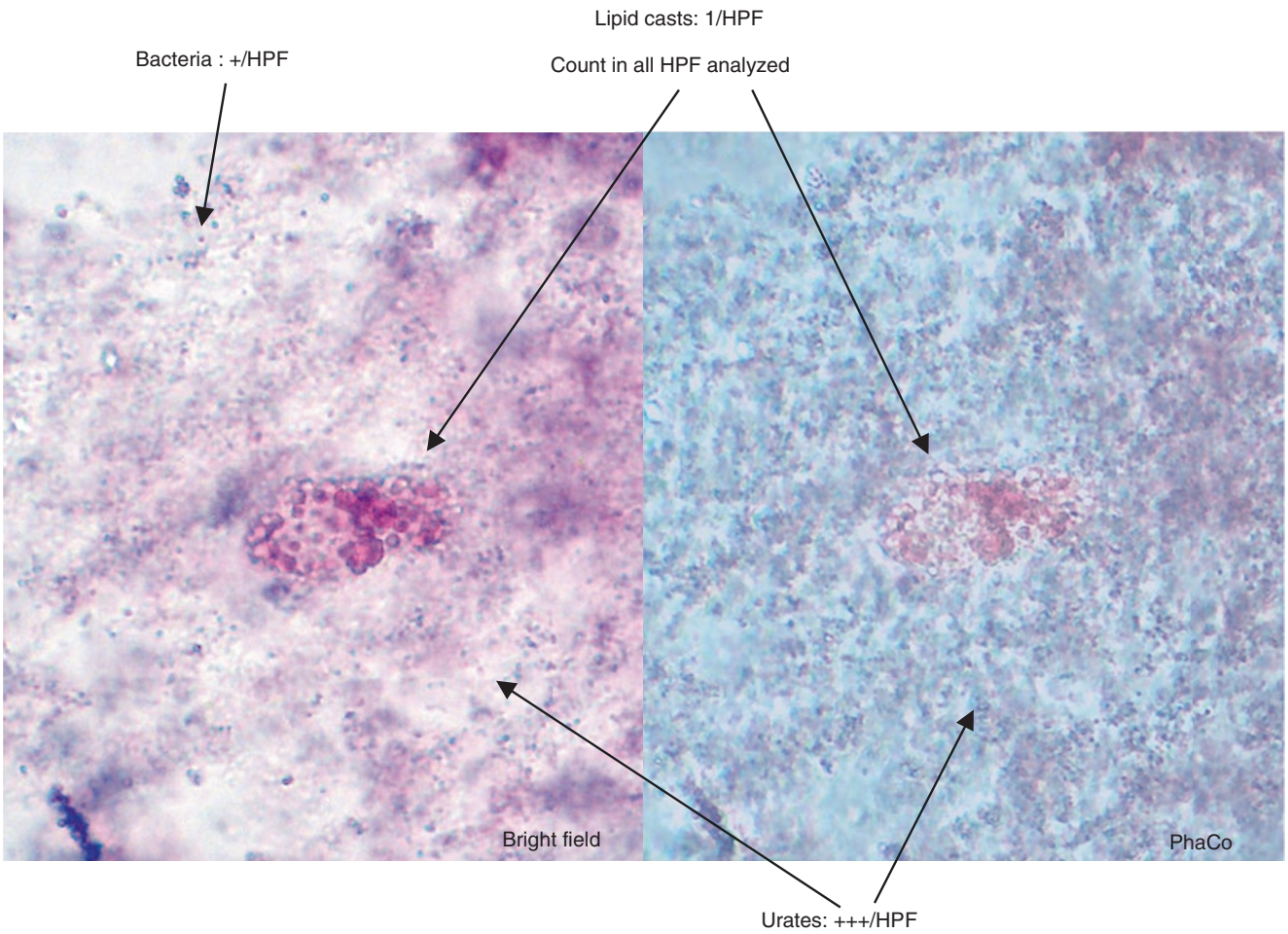


Fig. 12.36 Crystalluria and lipid casts: Sudan IV staining

12.3.22 Crystalluria

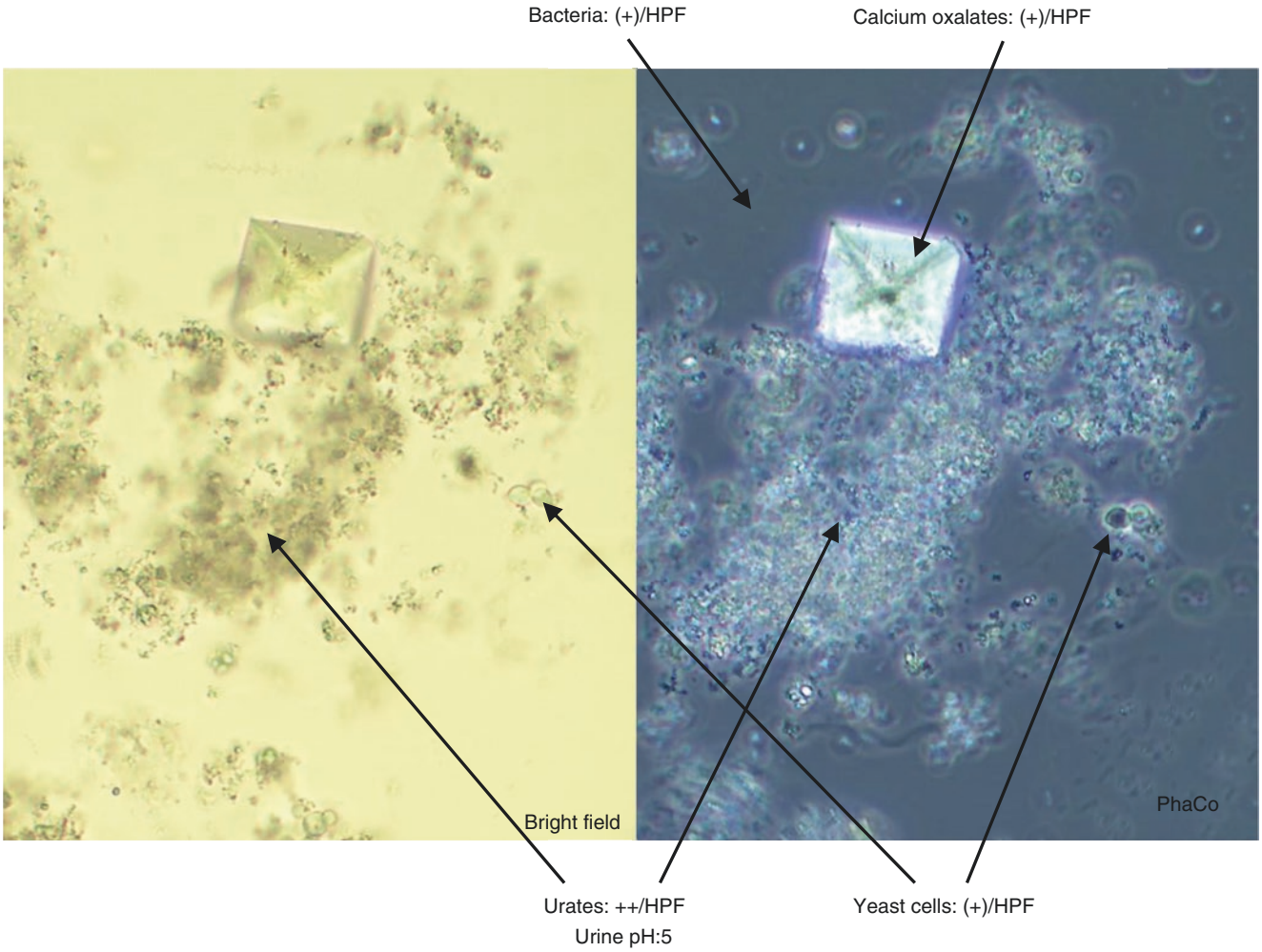


Fig. 12.37 Crystalluria I (calcium oxalates and urates)

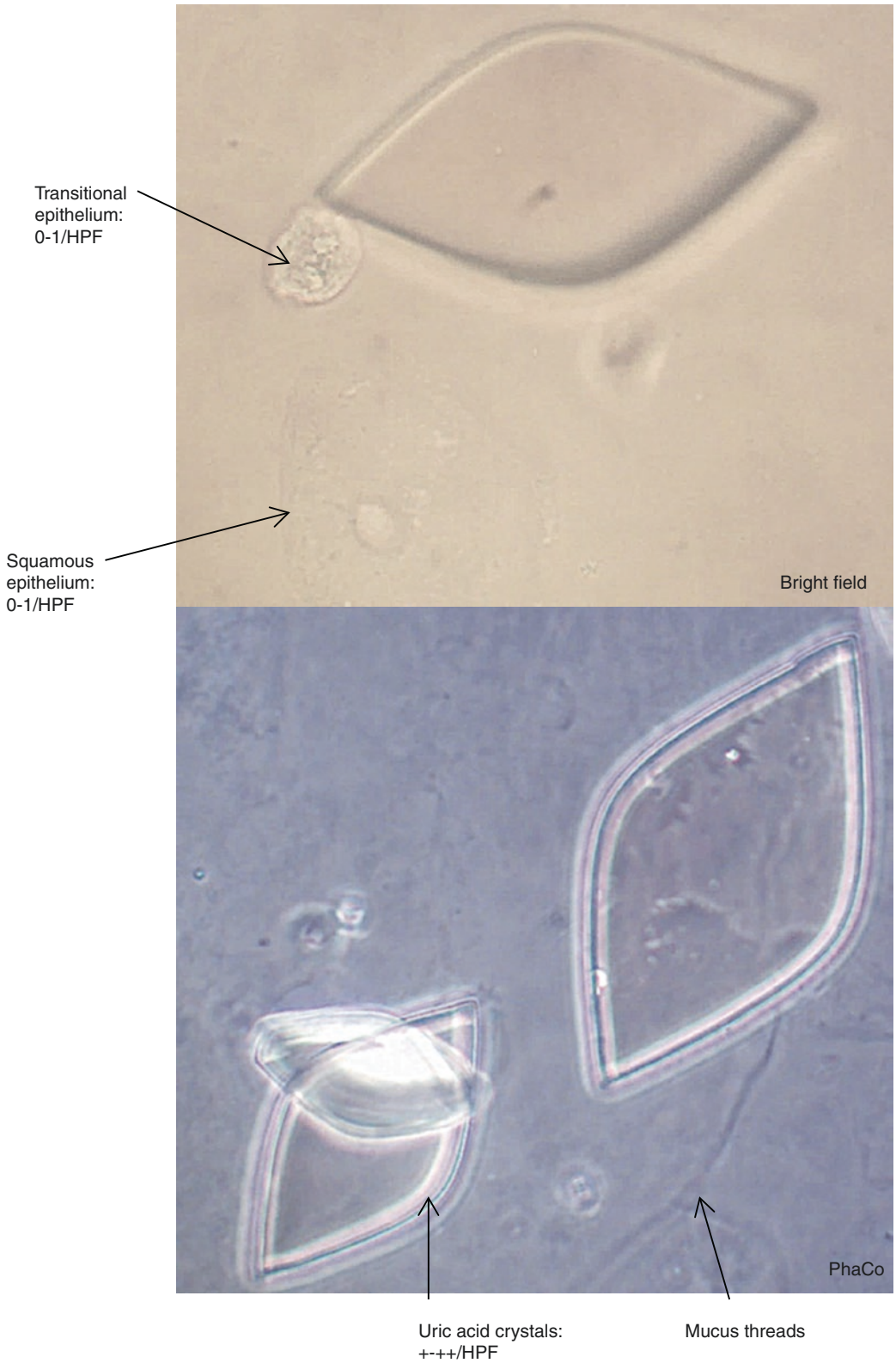
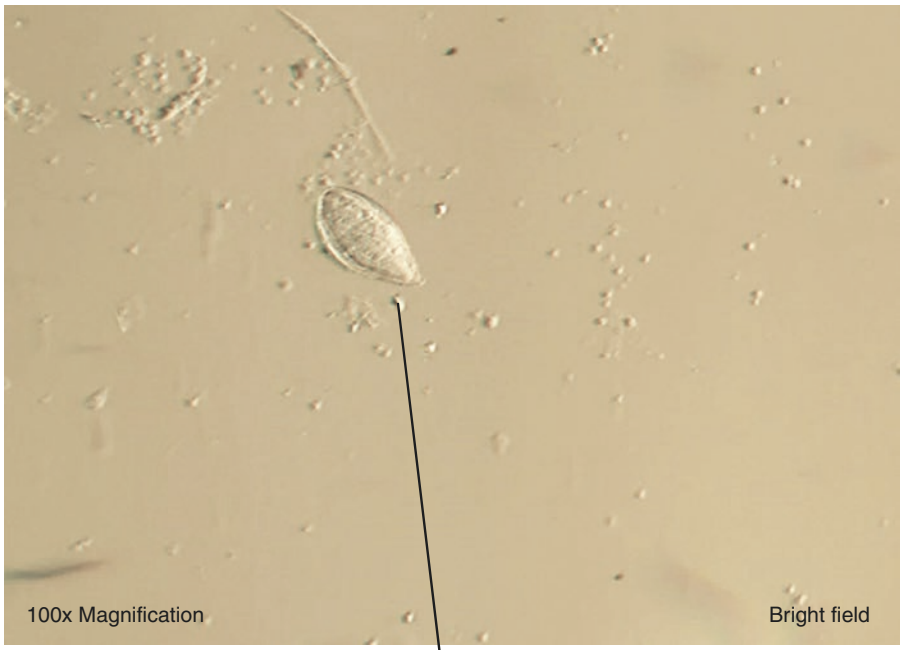
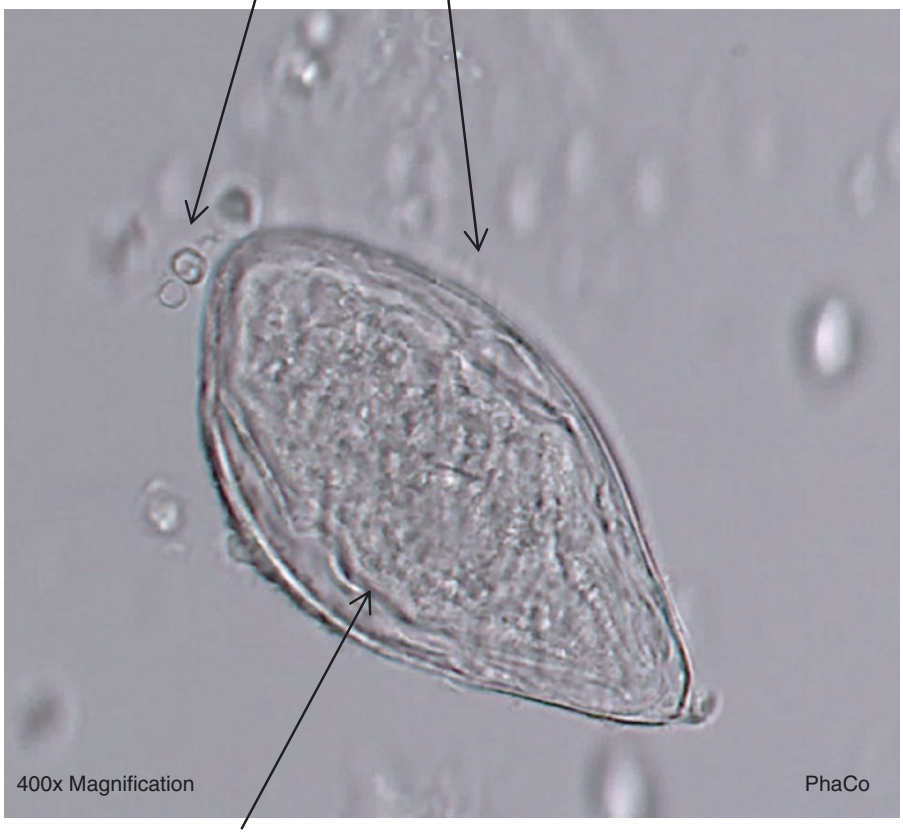


Fig. 12.38 Crystalluria II (uric acid crystals, diamond or rhombic shape)

12.3.23 *Schistosoma haematobium* Egg and Eumorphic Hematuria



Eumorphic erythrocytes: 5–15/HPF (not all eumorphic erythrocytes can be identified in this microscopic plane)



Schistosoma haematobium egg: (+)/HPF

Tip: Schistosoma haematobium eggs can be seen even at 100x overview magnification.

Fig. 12.39 *Schistosoma haematobium* egg and eumorphic hematuria

12.4 Diagnosis

12.4.1 Exercises in the diagnosis of microscopic urinary sediment images

Content of Findings

- The basic finding is an essential basic component of urinary sediment findings. Semiquantitative statements on eumorphic erythrocytes, leukocytes, squamous epithelial cells, and bacteria are formulated here.
- The basic findings are supplemented by the semiquantitative data from all other microscopically analyzed urinary sediment constituents, as well as a summation of the respective casts recorded in 20 visual fields.
- If there are dysmorphic erythrocytes/acanthocytes, their percentage is determined by counting eumorphic and dysmorphic erythrocytes and acanthocytes per 100 erythrocytes.
- Atypical-appearing cells that cannot be further classified are given a cell description.
- Information on artifacts and mucus threads can be omitted.

The following enables the reader to practice diagnosing microscopic urinary sediment images.

12.4.2 Findings Sheet: Urine Status

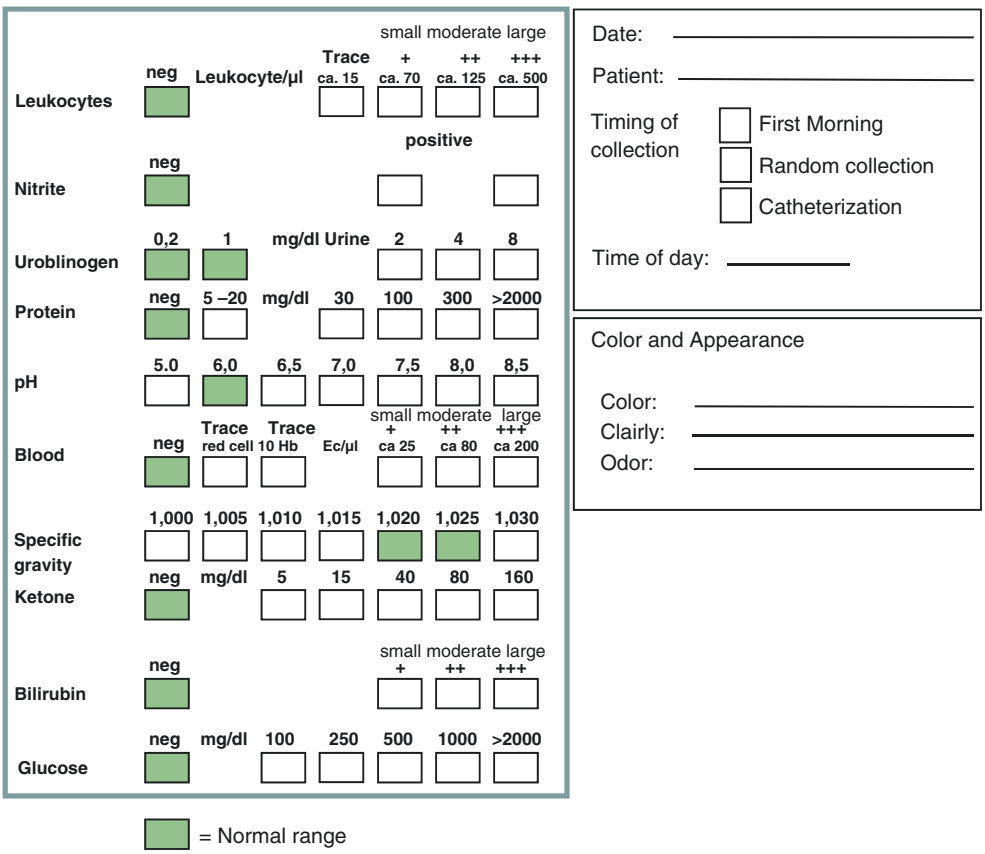


Fig. 12.40 Findings sheet: urine chemistry using test strips (urine test strip sample with kind permission from Siemens Healthcare Diagnostics)

Fig. 12.41 Findings sheet: urine sediment with reference to basic findings

Microscopic Examinaton of Urinary Sediment

Cells	HPF	
Erythrocytes - eumorphic		
Erythrocytes - dysmorphic		
Acanthocytes		
Leukocytes		
Histiocytes		
Squamous Epithelial Cells		
Transitional Epithelial Cells		
Deep Transitional Epi. Cells		
Renal Tub. Epithelial Cells		
Oval Fat Bodies		
Atypical Cells *		
Spermatozoa		

Percentage	
%	
%	
%	

Crystals:	HPF	
Uric Acid		
Urates		
Ammonium Urates		
Amorphous Phosphates		
Triple Phosphates		
Calcium Oxalates		
Calcium Phosphates		
Cholesterol		
Cystine		
Tyrosine		
Leucine		
Bilirubin		
Drug Crystals		
Free fat droplets		
Artifacts		

Casts	aHPF	
Hyaline		
Granular		
Waxy		
Epithelial Cell		
Erythrocytes		
Leukocytes		
Fatty		
Fat Bodies		
Hemoglobin-,Myoglobin		
Microorganisms	HPF	
Bacteria		
Yeast		
Parasites**		

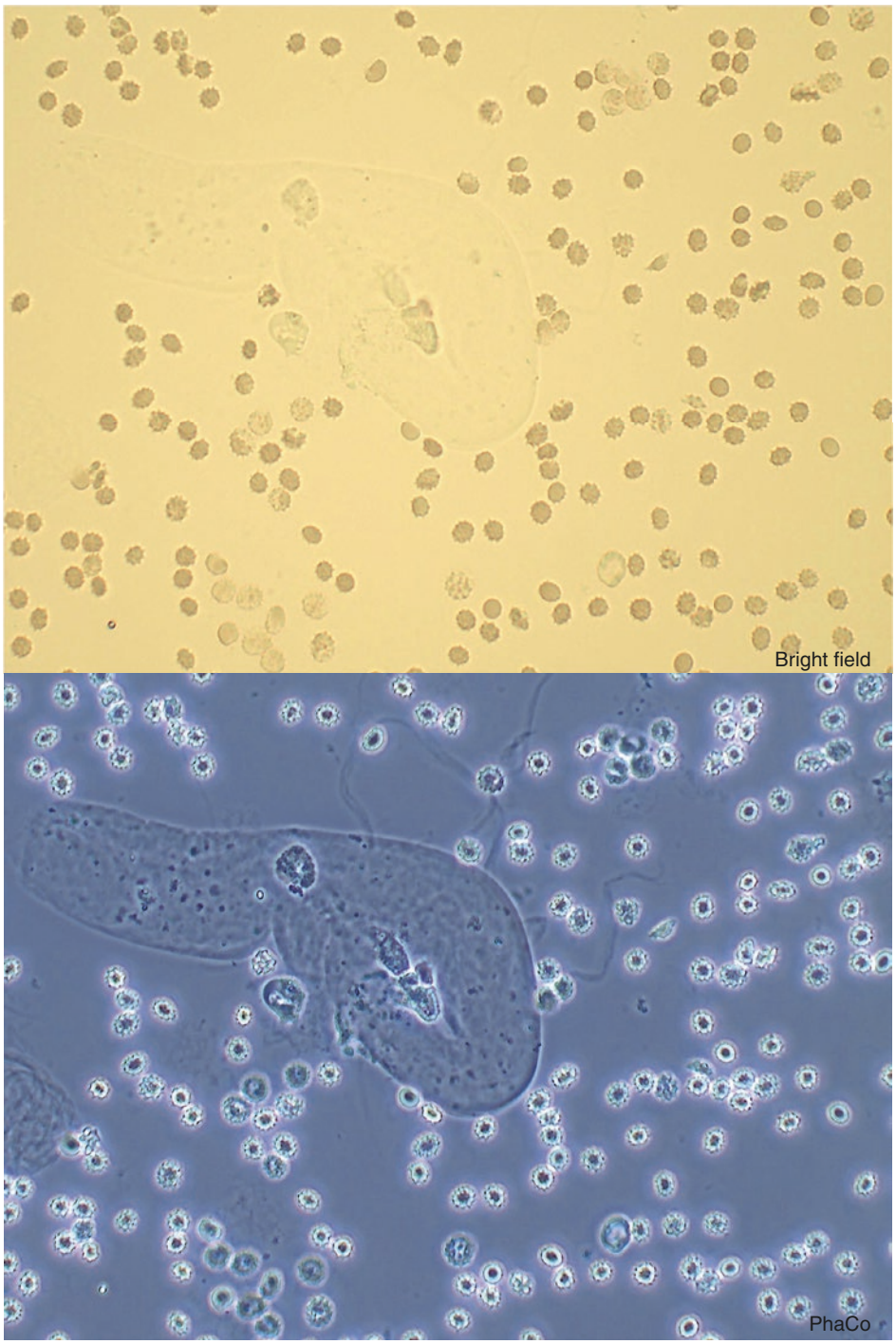
* Cell Description
Single/group/anisocytosis
Nucleus–cytoplasm ratio
Cell size/nucleus size
Nuclear structure/nuclear chromatin
Nucleoli
Granulation
Other inclusions

** *Schistosoma haematobium eggs, Trichomonas vaginalis, EnterobiusVermicularis* eggs

= Basic findings

HPF = High power field aHPF= Count in all HPF analyzed

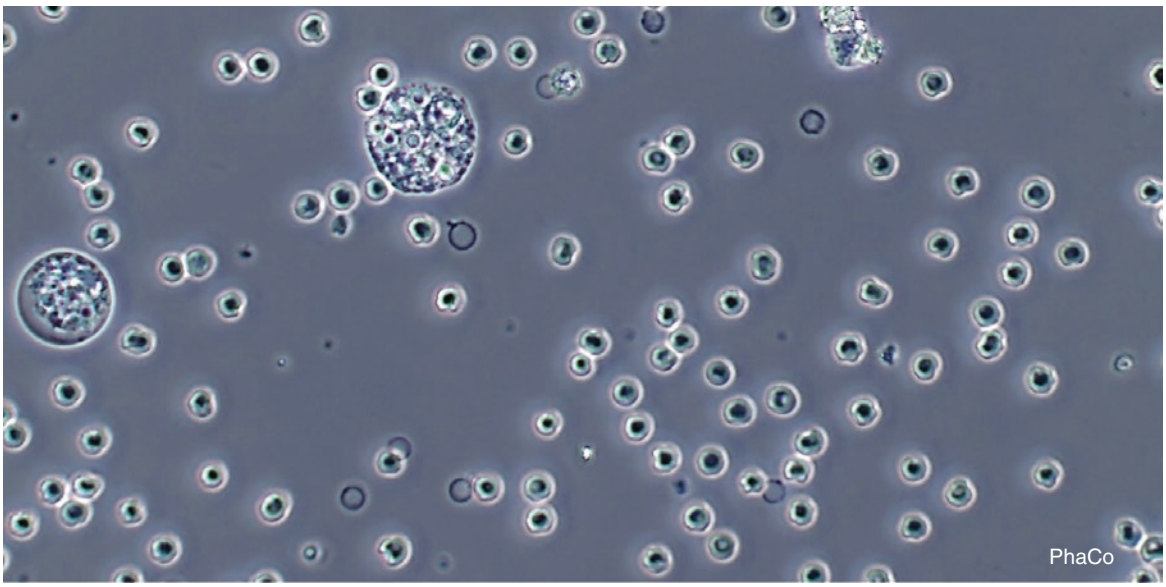
12.4.3 Eumorphic Hematuria (Thorn-Apple) with Fine Granular Cast



EumEc:	> 50	/HPF	SqEc:	0–1	/HPF
DysEc:	-	/HPF	Bact:	(+)	/HPF
Lc:	0–1	/HPF	GranCa:	1	/aHPF

Fig. 12.42 Eumorphic hematuria (thorn-apple) with fine granular cast

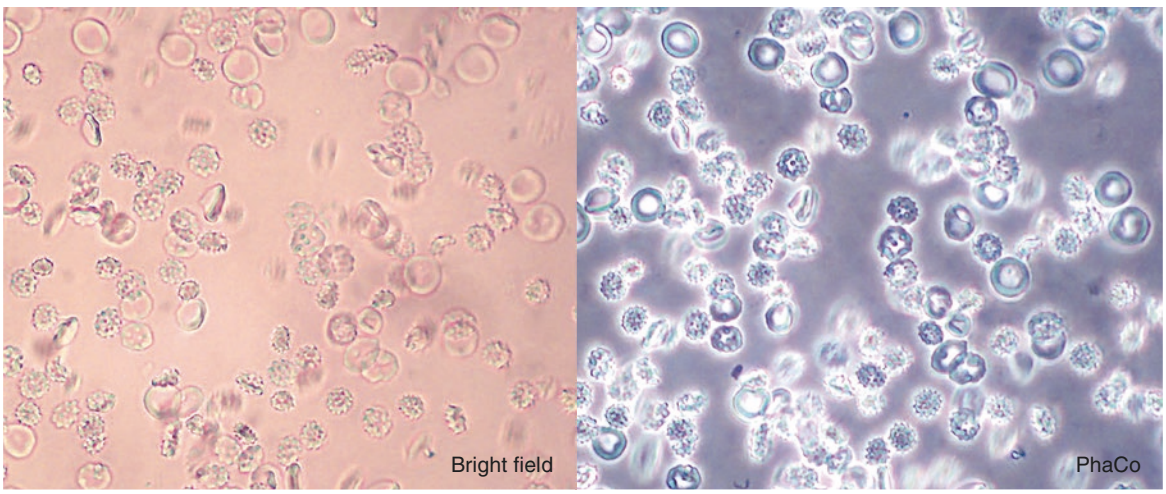
12.4.4 Eumorphic Hematuria with Histiocytes



EumEc:	> 50	/HPF	SqEc:	0–1	/HPF
DysEc:	-	/HPF	Bact:	(+)	/HPF
Lc:	0–1	/HPF	Histiocytes:	1–4	/HPF

Fig. 12.43 Eumorphic hematuria with histiocytes

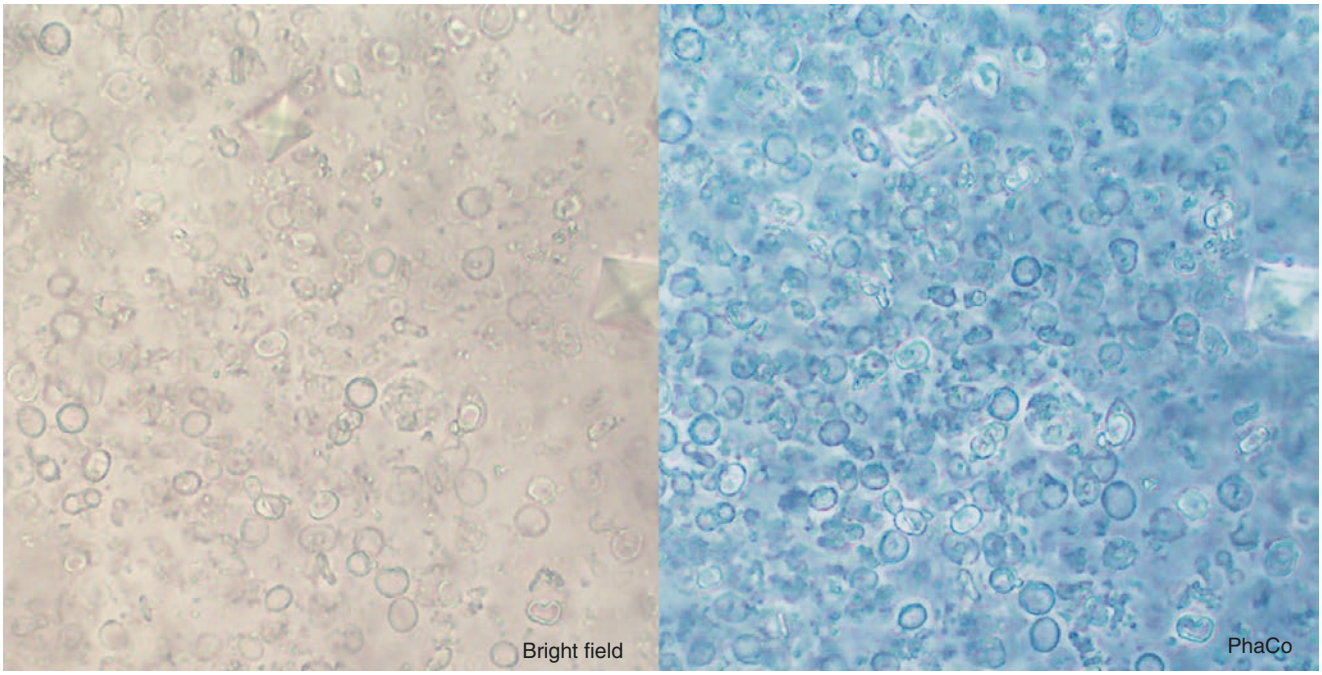
12.4.5 Eumorphic Hematuria



EumEc:	> 50	/HPF	SqEc:	0–1	/HPF
DysEc:	–	/HPF	Bact:	(+)	/HPF
Lc:	0–1	/HPF			

Fig. 12.44 Eumorphic hematuria

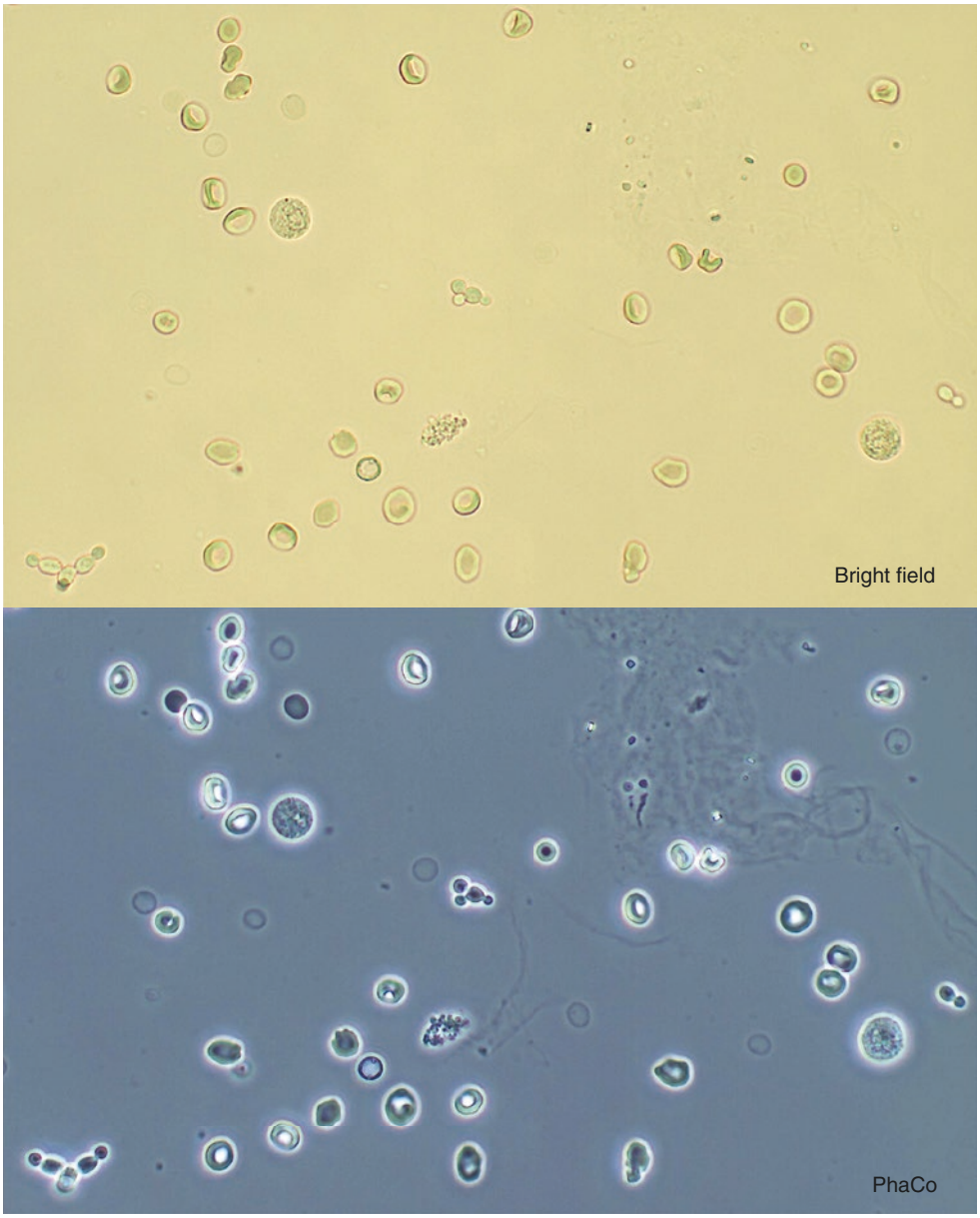
12.4.6 Eumorphic Hematuria and Crystalluria



EumEc:	> 50	/HPF	SqEc:	0–1	/HPF
DysEc:	–	/HPF	Bact:	+	/HPF
Lc:	0–1	/HPF	Ca-oxalates:	(+)–+	/HPF

Fig. 12.45 Eumorphic hematuria and crystalluria (calcium oxalates, square/envelope-shaped)

12.4.7 Eumorphic Hematuria and Yeast Cells



EumEc:	15–50	/HPF	SqEc:	0–1	/HPF
DysEc:		/HPF	Bact:	(+)	/HPF
Lc:	1–4	/HPF	Yeast cells:	(+)	/HPF

Tip: The morphologically varied erythrocytes (erythrocyte ghosts, biconcave erythrocytes) are striking. The low-contrast erythrocyte ghosts cannot be differentiated in bright-field mode.

Fig. 12.46 Eumorphic hematuria and yeast cells

12.4.8 Dysmorphic Hematuria

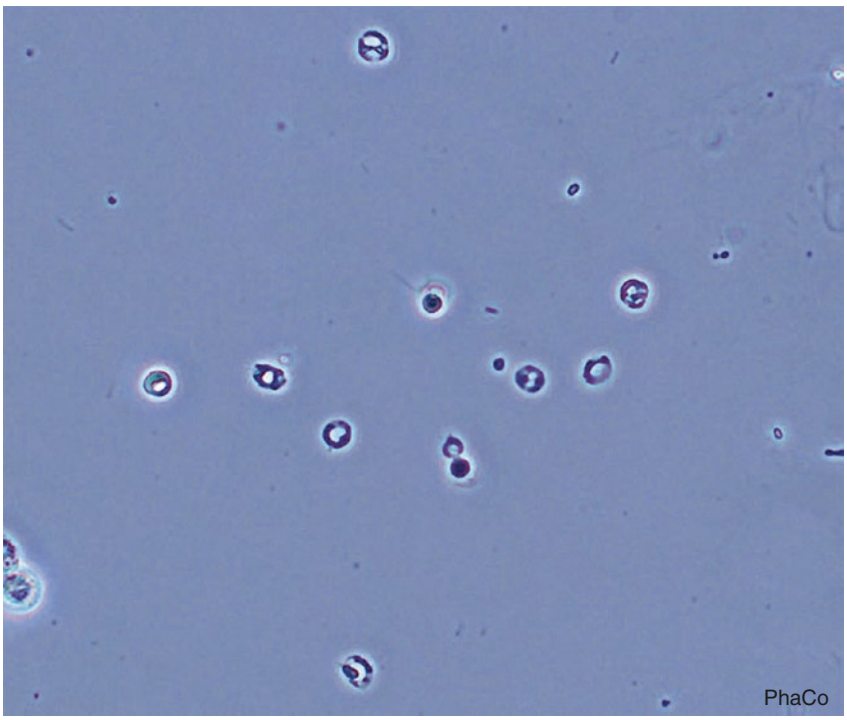


Fig. 12.47 Dysmorphic hematuria → suspected renal hematuria

EumEc:	0-1	/HPF	SqEc:	0-1	/HPF	EumEc:	10	%
DysEc:	5-15	/HPF	Bact:	+	/HPF	DysEc:	80	%
Lc:	0-1	/HPF				Acantho:	10	%

100 Ec

Count the number of eumorphic erythrocytes, dysmorphic erythrocytes, and acanthocytes per 100 erythrocytes and determine the percentage.

12.4.9 Dysmorphic Hematuria with Erythrocyte Cast

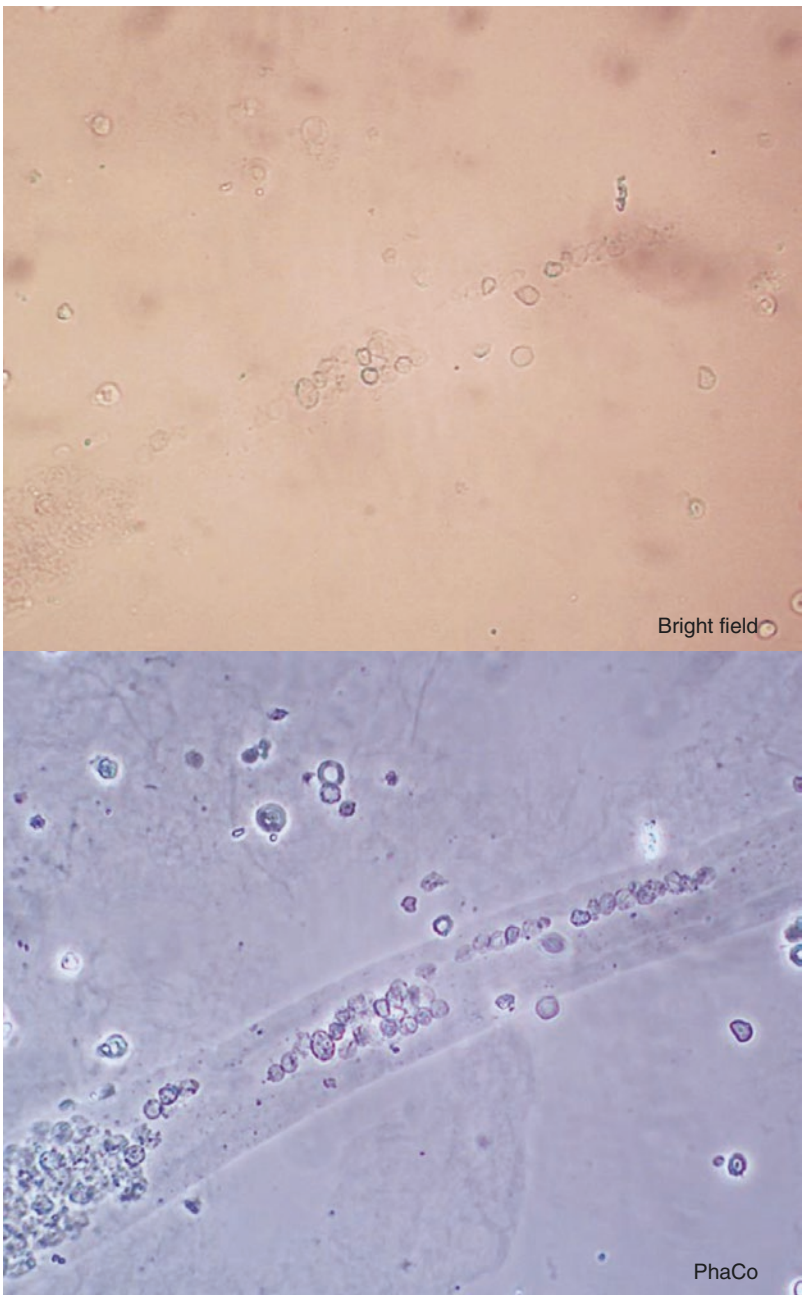


Fig. 12.48 Dysmorphic hematuria with erythrocyte cast → suspected renal hematuria

EumEc:	0-1	/HPF	Plepi:	0-1	/HPF	EumEc:	17	%
DysEc:	5-15	/HPF	Bakt:	(+)	/HPF	DysEc:	75	%
Lc:	0-1	/HPF	EcCa:	1	/aHPF	Acantho:	8	%

100 Ec

Count the number of eumorphic erythrocytes, dysmorphic erythrocytes, and acanthocytes per 100 erythrocytes and determine the percentage.

12.4.10 Erythrocyte Casts

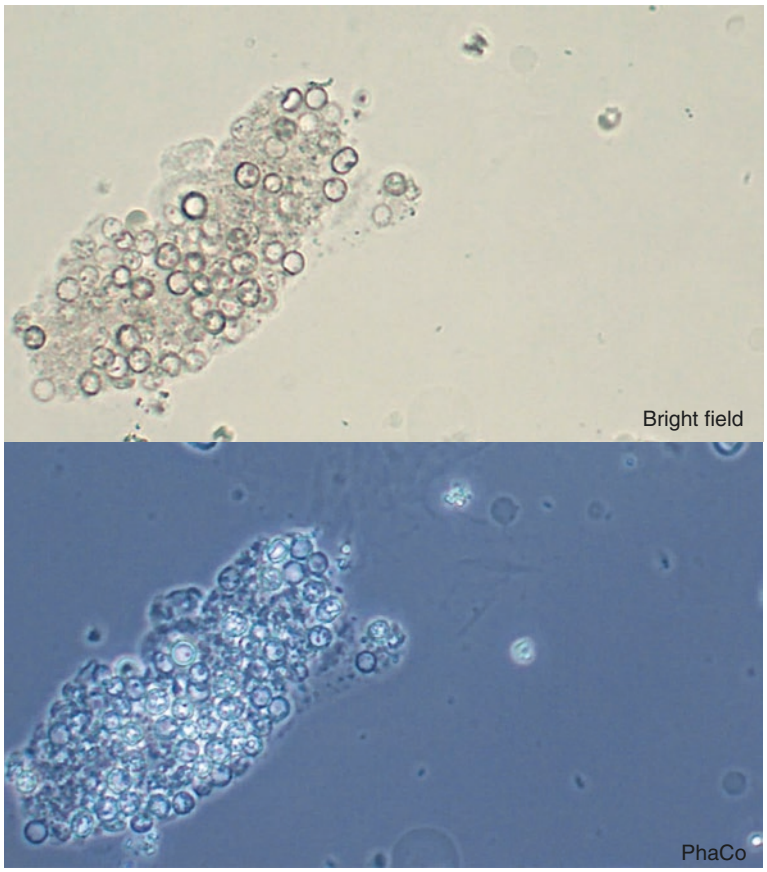
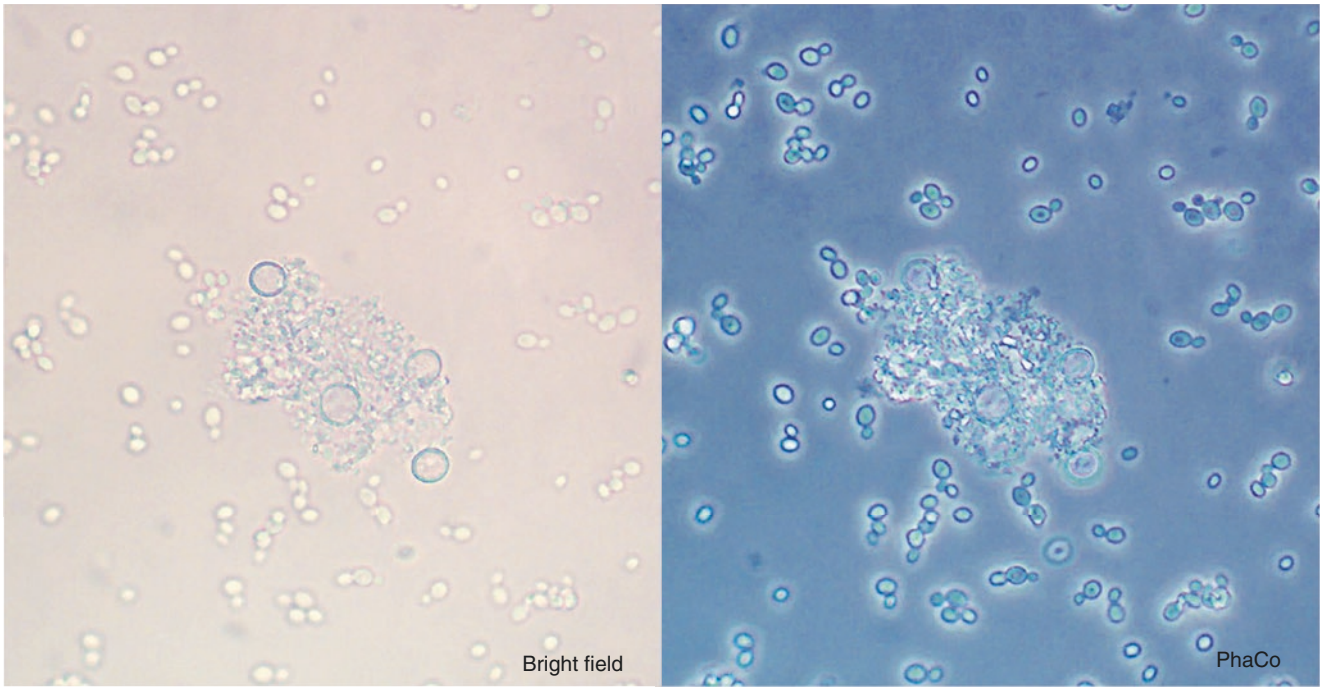


Fig. 12.49 Erythrocyte casts → suspected renal hematuria

EumEc:	0–1	/HPF	SqEc:	0–1	/HPF	EumEc: 20	%	100 Ec
DysEc:	1–4	/HPF	Bact:	(+)	/HPF	DysEc: 78	%	
Lc:	0–1	/HPF	EcCa:	4	/aHPF	Acantho: 2	%	

Count the number of eumorphic erythrocytes, dysmorphic erythrocytes, and acanthocytes per 100 erythrocytes and determine the percentage.

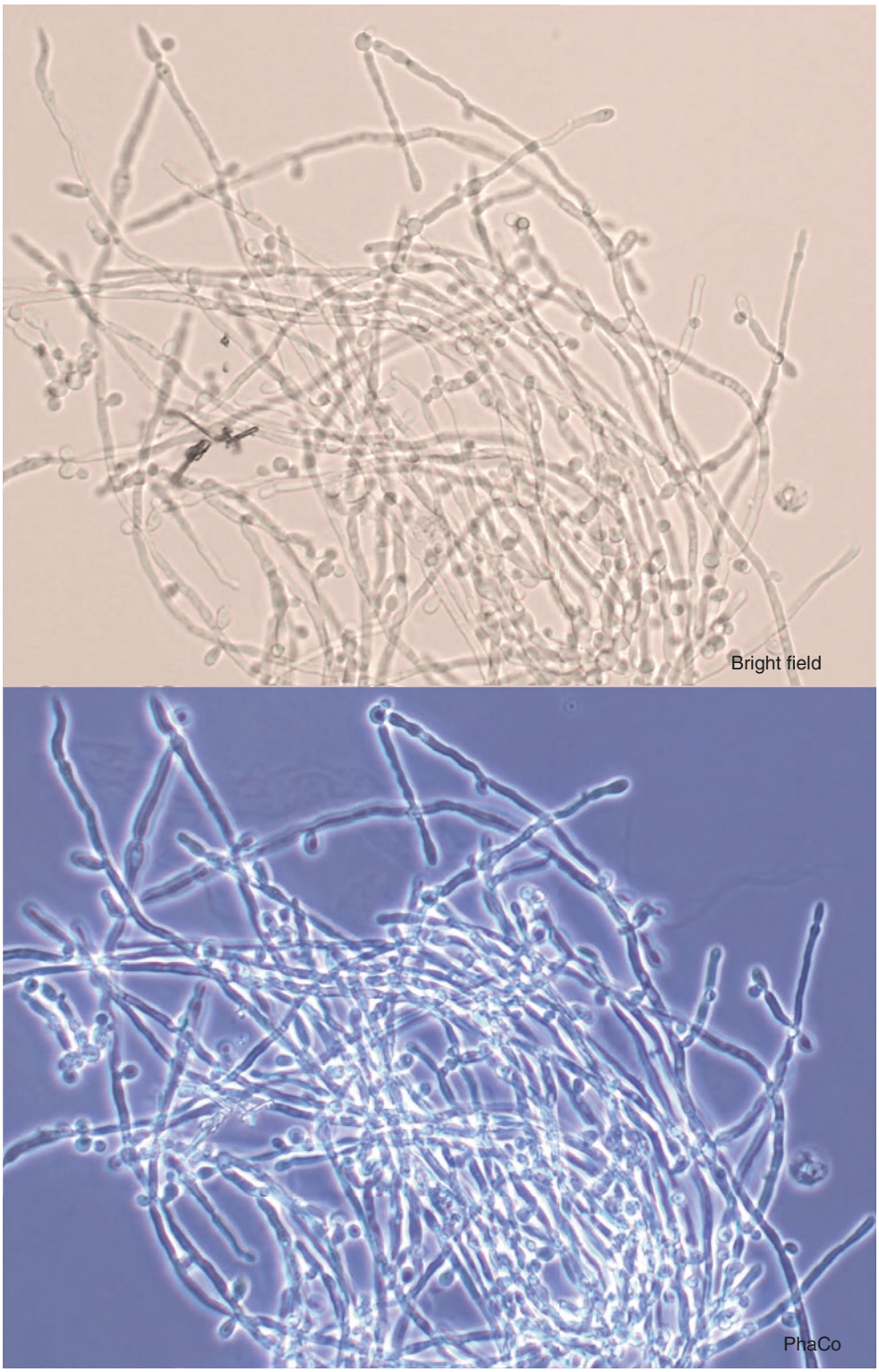
12.4.11 Yeast Cells with Chlamydospores



EumEc:	0–1	/HPF	SqEc:	0–1	/HPF
DysEc:		/HPF	Bact:	(+)	/HPF
Lc:	0–1	/HPF	Yeast cells: with four chalmydospores	++	/HPF

Fig. 12.50 Yeast cells with chlamydospores

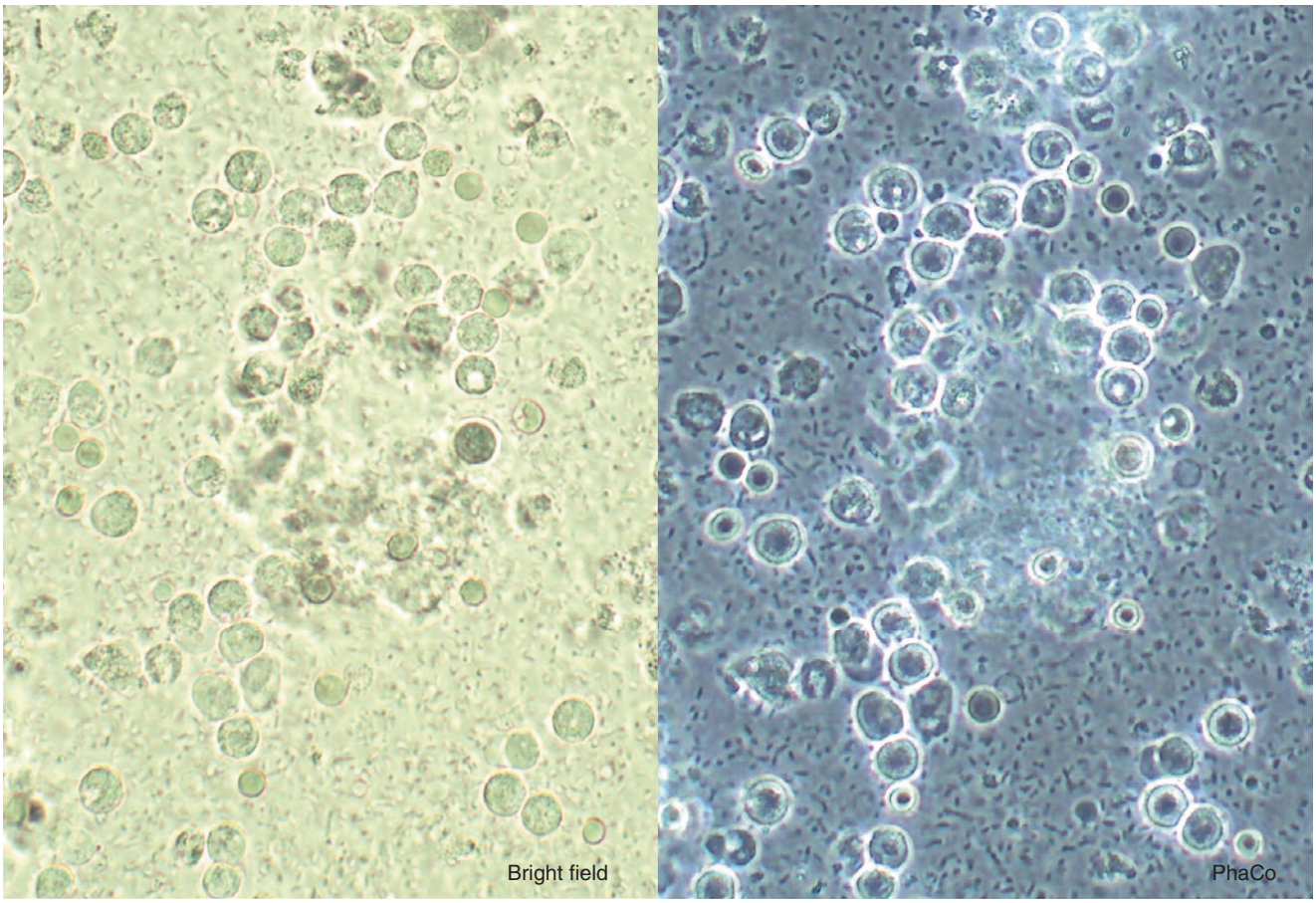
12.4.12 Yeast Cells and Fungal Hyphae



EumEc:	0–1	/HPF	SqEc:	0–1	/HPF
DysEc:		/HPF	Bact:	(+)	/HPF
Lc:	0–1	/HPF	Yeast cells/fungal hyphae:	++	/HPF

Fig. 12.51 Yeast cells and fungal hyphae

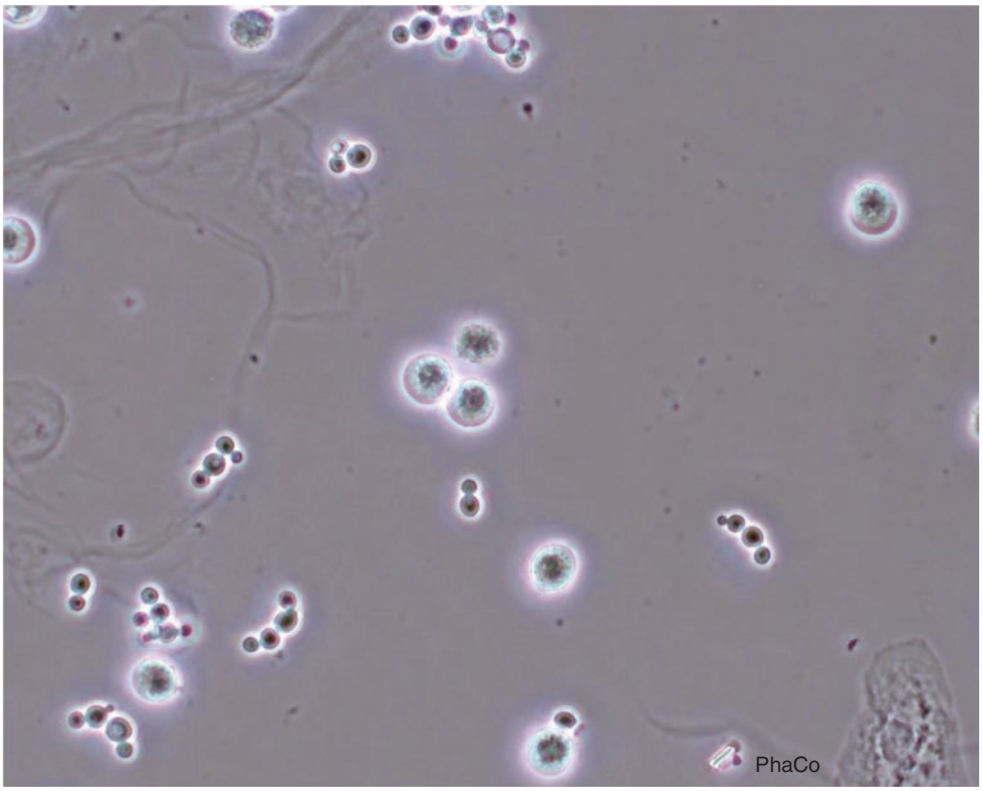
12.4.13 Leukocyturia with Bacteriuria and Eumorphic Hematuria



EumEc:	5–15	/HPF	SqEc:	0–1	/HPF
DysEc:		/HPF	Bact:	+++	/HPF
Lc:	15–50	/HPF			

Fig. 12.52 Leukocyturia, bacteriuria, and eumorphic hematuria → suspected bacterial urinary tract infection accompanied by bleeding

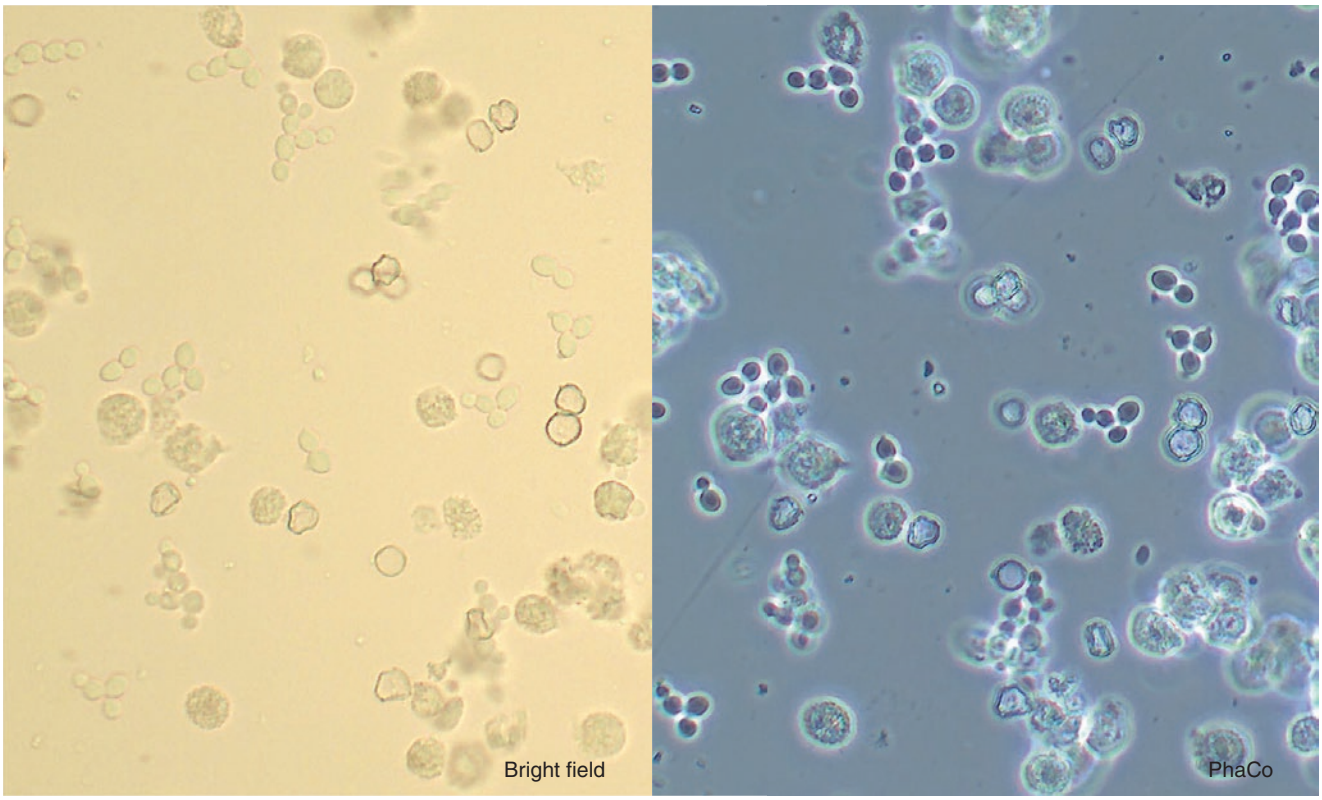
12.4.14 Leukocyturia and Yeast Cells



EumEc:	0–1	/HPF	SqEc:	0–1	/HPF
DysEc:		/HPF	Bact:	(+)	/HPF
Lc:	5–15	/HPF	Yeast cells:	(+)	/HPF

Fig. 12.53 Leukocyturia and yeast cells → suspected yeast infection

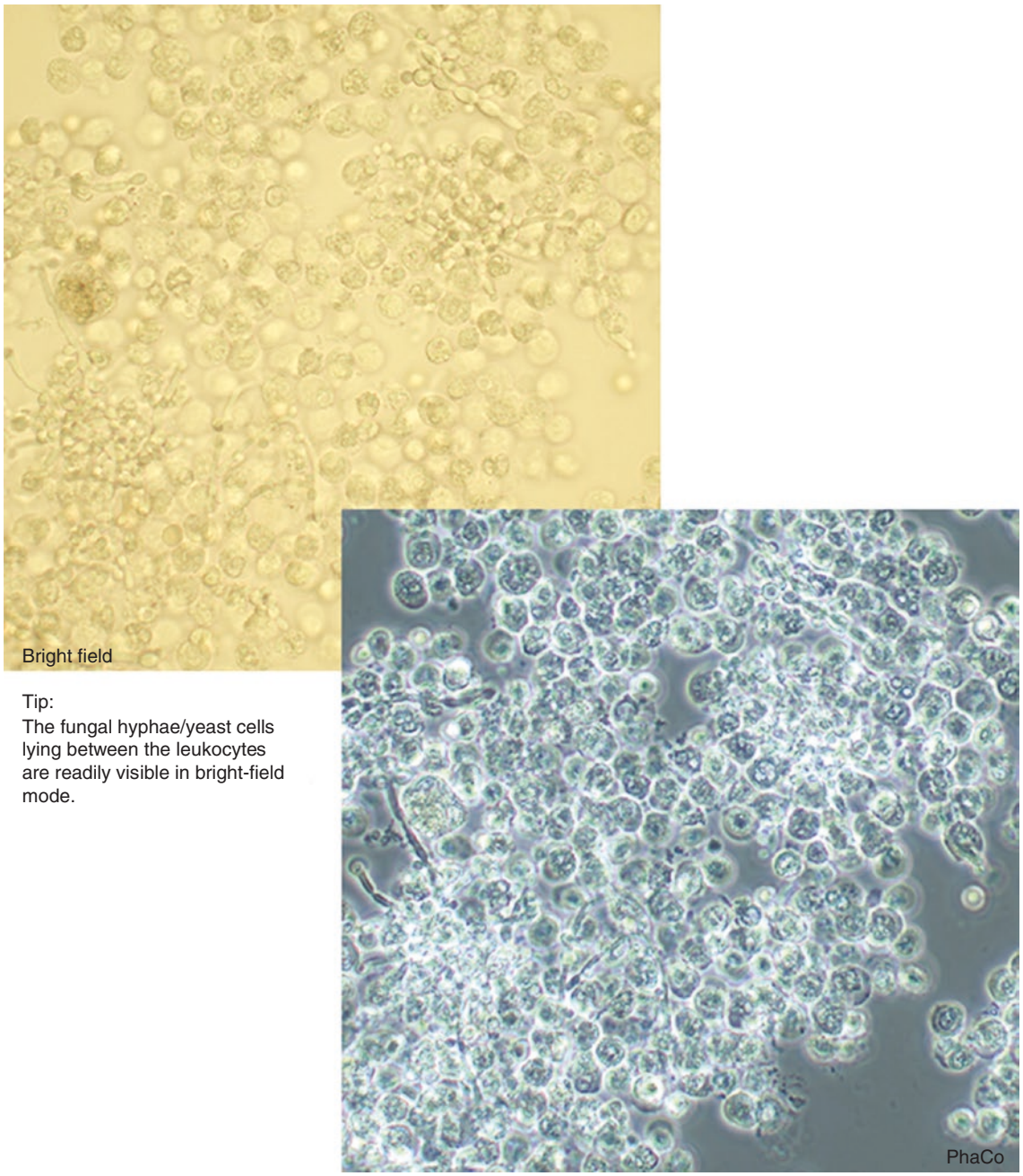
12.4.15 Leukocyturia with Yeast Cells and Eumorphic Hematuria



EumEc: 5–15 (Ec ghosts)	/HPF	SqEc: 0–1	/HPF
DysEc:	/HPF	Bact: (+)	/HPF
Lc: 15–50	/HPF	Yeast cells: +-++	/HPF

Fig. 12.54 Leukocyturia with yeast cells and eumorphic hematuria → suspected yeast infection accompanied by bleeding

12.4.16 Leukocyturia with Fungal Hyphae and Yeast Cells

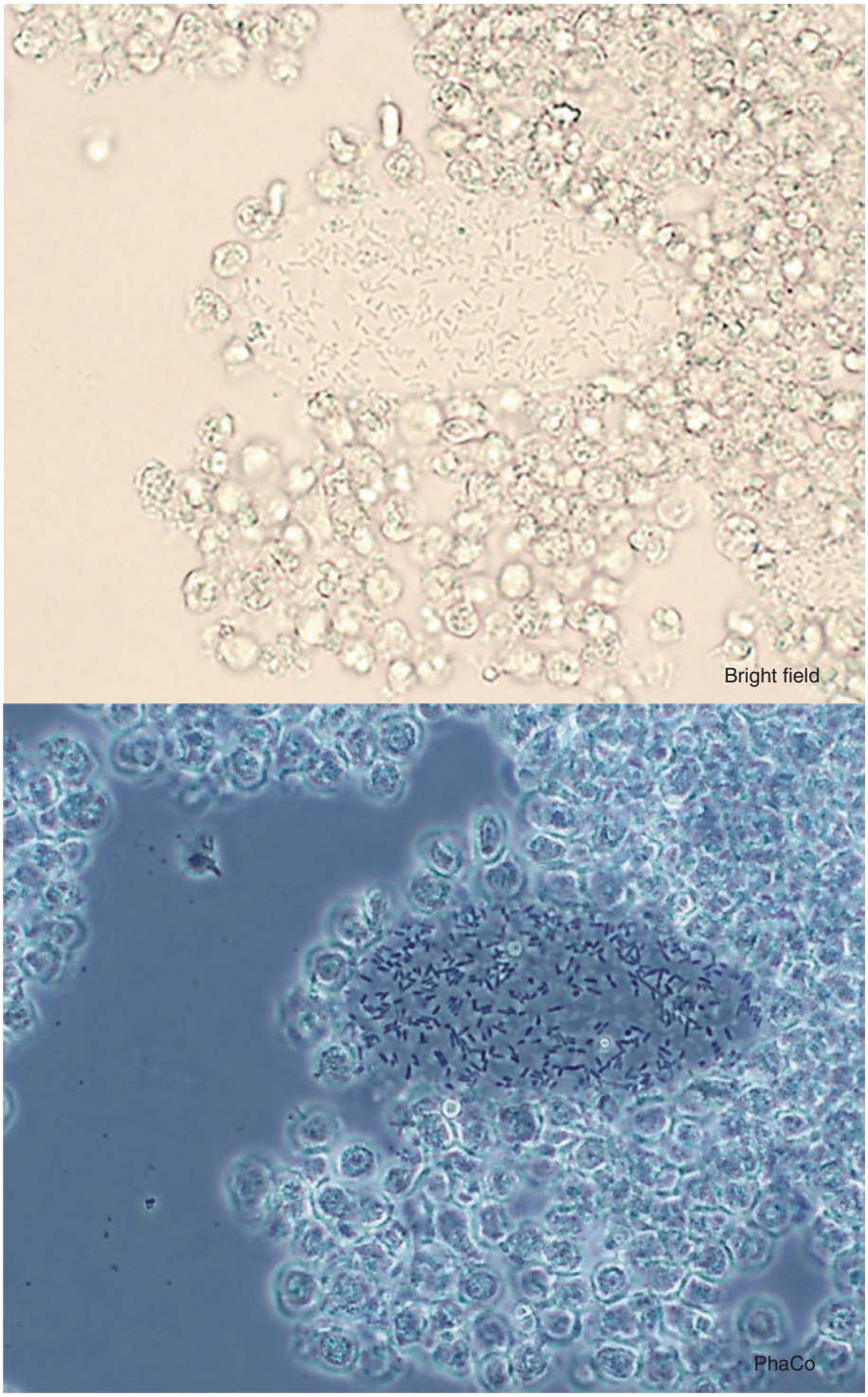


Tip:
The fungal hyphae/yeast cells lying between the leukocytes are readily visible in bright-field mode.

EumEc:	1–4	/HPF	SqEc:	0–1	/HPF
DysEc:		/HPF	Bact:	(+)	/HPF
Lc:	>50	/HPF	Yeast cells and: fungal hyphae	+-++	/HPF

Fig. 12.55 Leukocyturia with fungal hyphae → suspected yeast infection

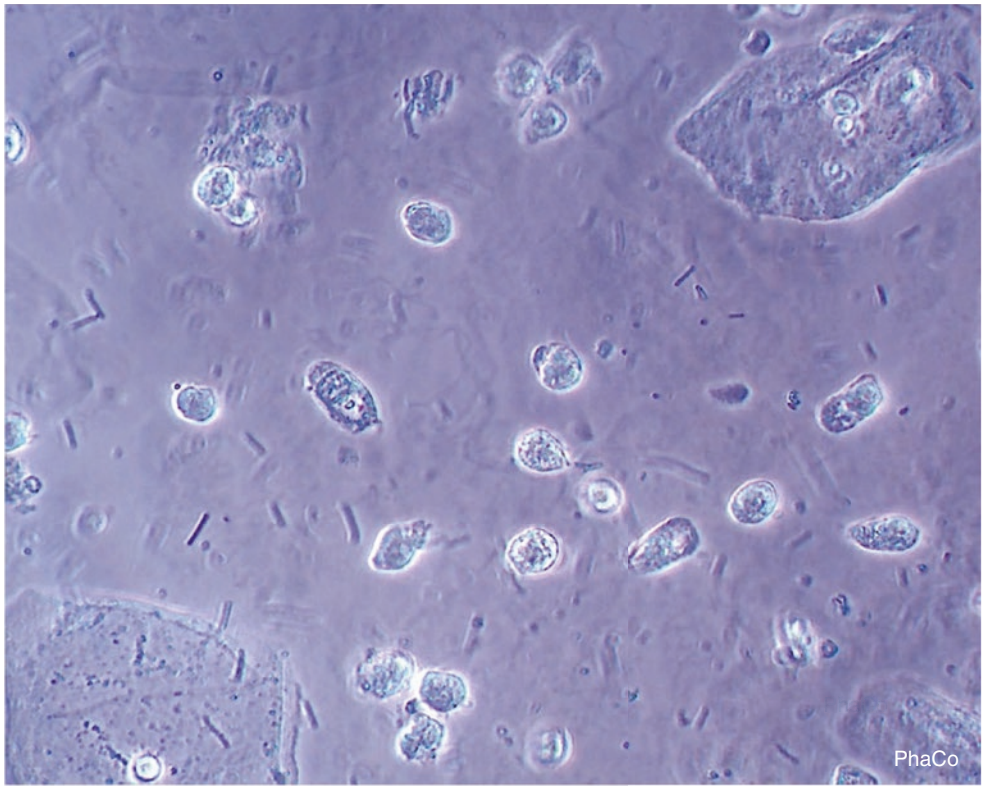
12.4.17 Leukocyturia with Bacterial Casts



EumEc:	0–1	/HPF	SqEc:	0–1	/HPF
DysEc:		/HPF	Bact:	++	/HPF
Lc:	>50	/HPF	Bacterial casts:	10	/aHPF

Fig. 12.56 Leukocyturia with bacterial casts → suspected severe bacterial urinary tract infection with renal involvement

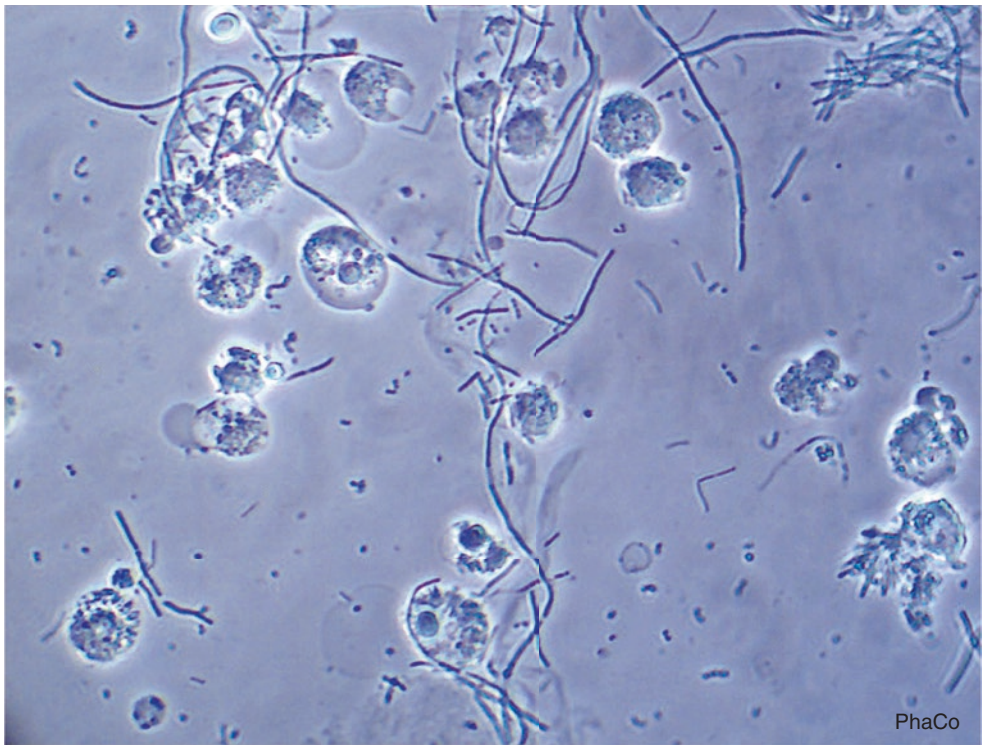
12.4.18 Leukocyturia and Bacteriuria with Deep Urothelial Cells



EumEc:	1–4	/HPF	SqEc:	1–4	/HPF
DysEc:		/HPF	Bact:	+-++	/HPF
Lc:	5–15	/HPF	Deep urothelial cells:	1–4	/HPF

Fig. 12.57 Leukocyturia and bacteriuria with deep urothelial cells

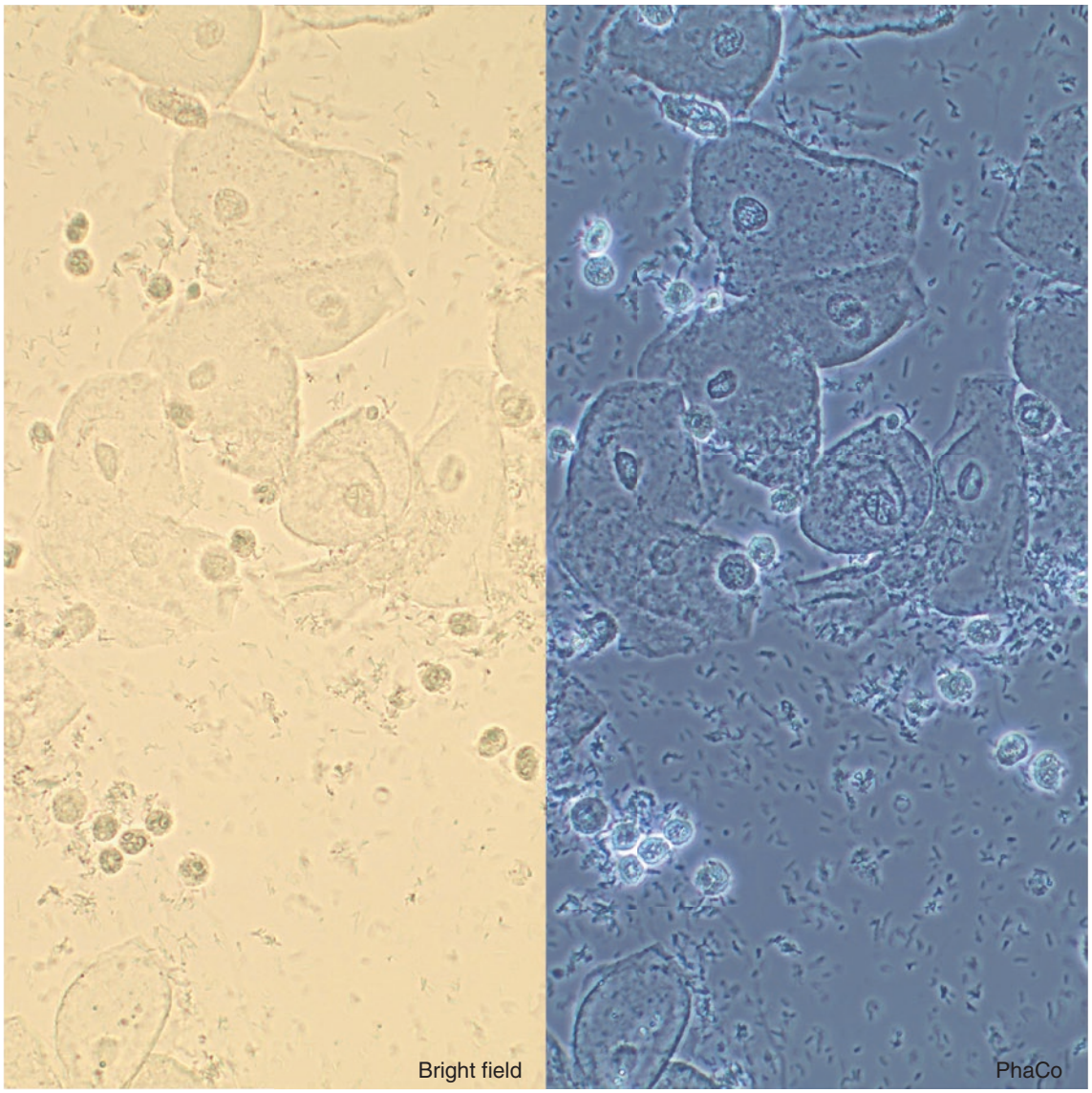
12.4.19 Leukocyturia and Bacteriuria: Old Urine Sample



EumEc:	1–4	/HPF	SqEc:	0–1	/HPF
DysEc:		/HPF	Bact:	+-++	/HPF
Age (!) Lc:	15–50	/HPF			

Fig. 12.58 Leukocyturia and bacteriuria: old urine sample

12.4.20 Suspected Pseudo-urinary Tract Infection

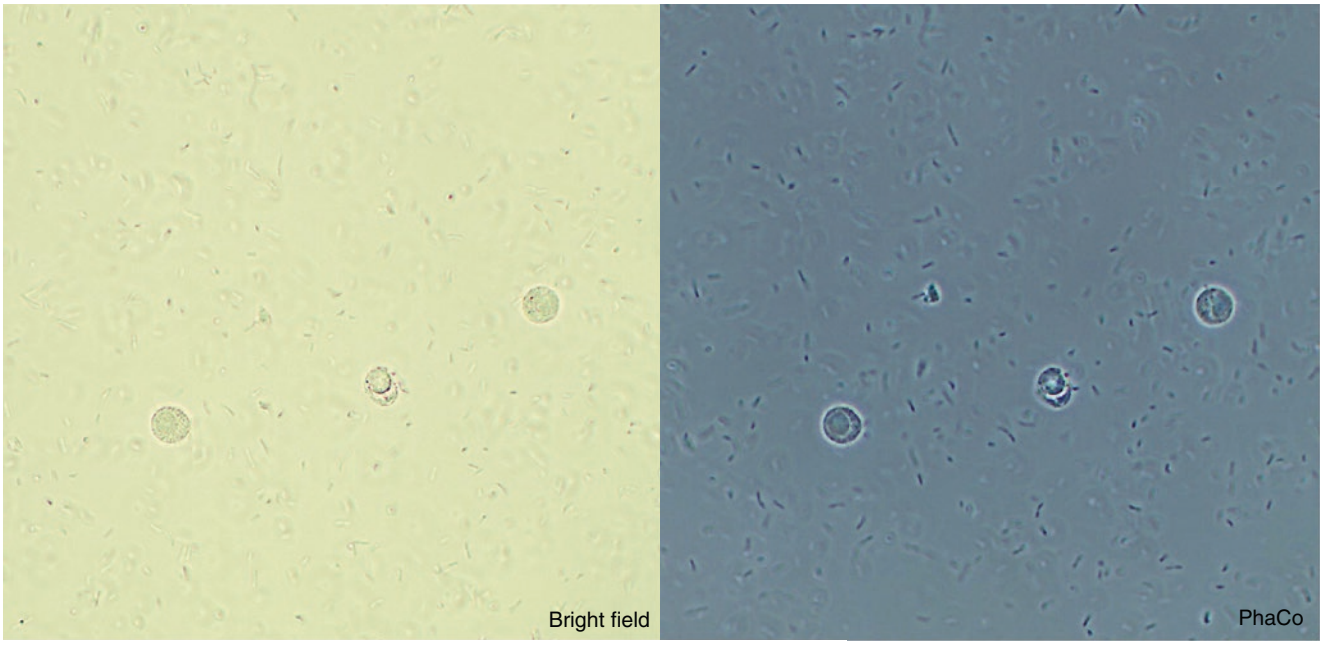


EumEc:	0–1	/HPF	SqEc:	5–15	/HPF
DysEc:		/HPF	Bact:	++	/HPF
Lc:	5–15	/HPF			

Tip: The increased presence of squamous epithelial cells (>7–8/HPF, 18 mm field number) in urine sediment indicates that the urine sample was not taken from the midstream. Therefore, the leukocytes and bacteria that are also increased here could originate from the outer genital tract. For this reason, the presence of a urinary tract infection is considered questionable.

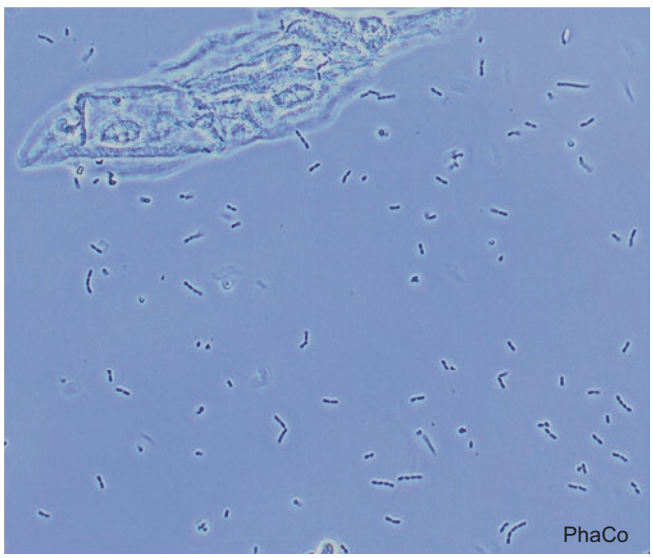
Fig. 12.59 Suspected pseudo-urinary tract infection

12.4.21 Bacteriuria



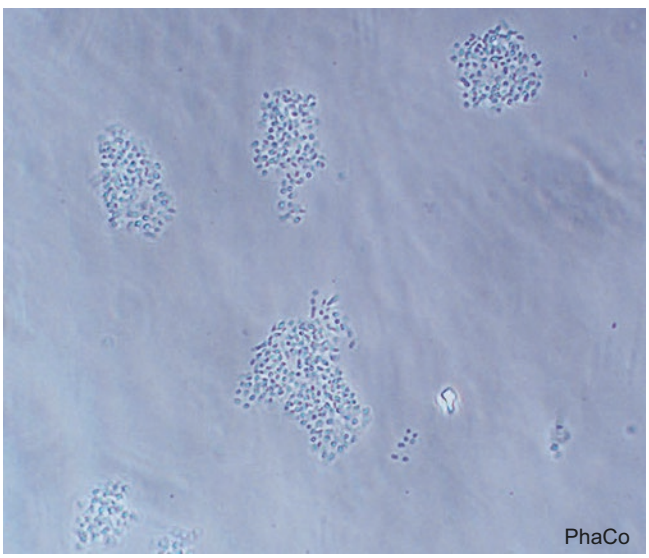
EumEc: 0–1	/HPF	SqEc: 0–1	/HPF
DysEc:	/HPF	Bact: ++-+++	/HPF
Lc: 1–4	/HPF		

Fig. 12.60 Bacteriuria I



EumEc: 0–1	/HPF	SqEc: 1–4	/HPF
DysEc:	/HPF	Bact: +–++	/HPF
Lc: 0–1	/HPF		

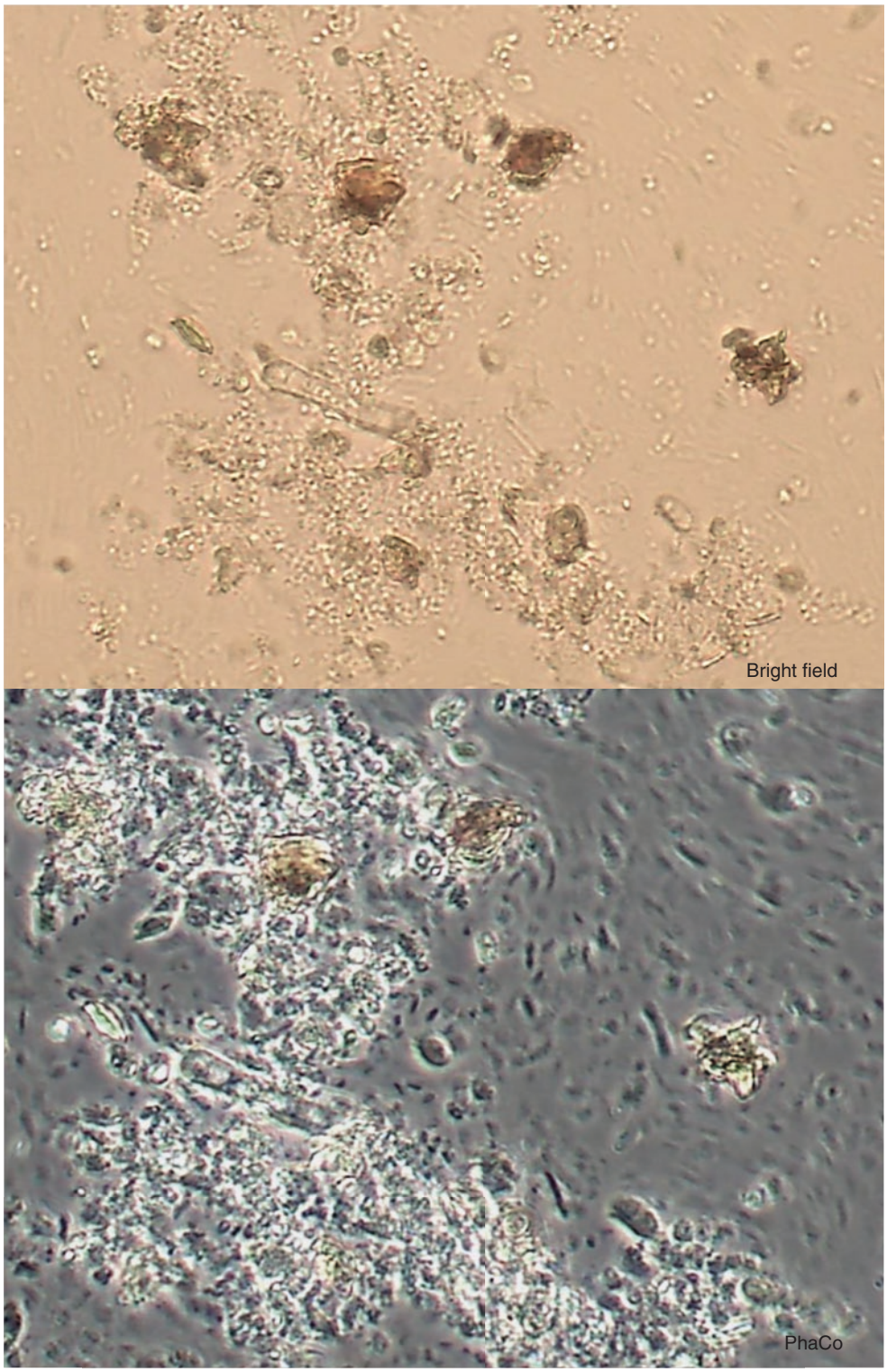
Fig. 12.61 Bacteriuria II



EumEc: 0–1	/HPF	SqEc: 0–1	/HPF
DysEc:	/HPF	Bact: +–++	/HPF
Lc: 0–1	/HPF		

Fig. 12.62 Bacteriuria III

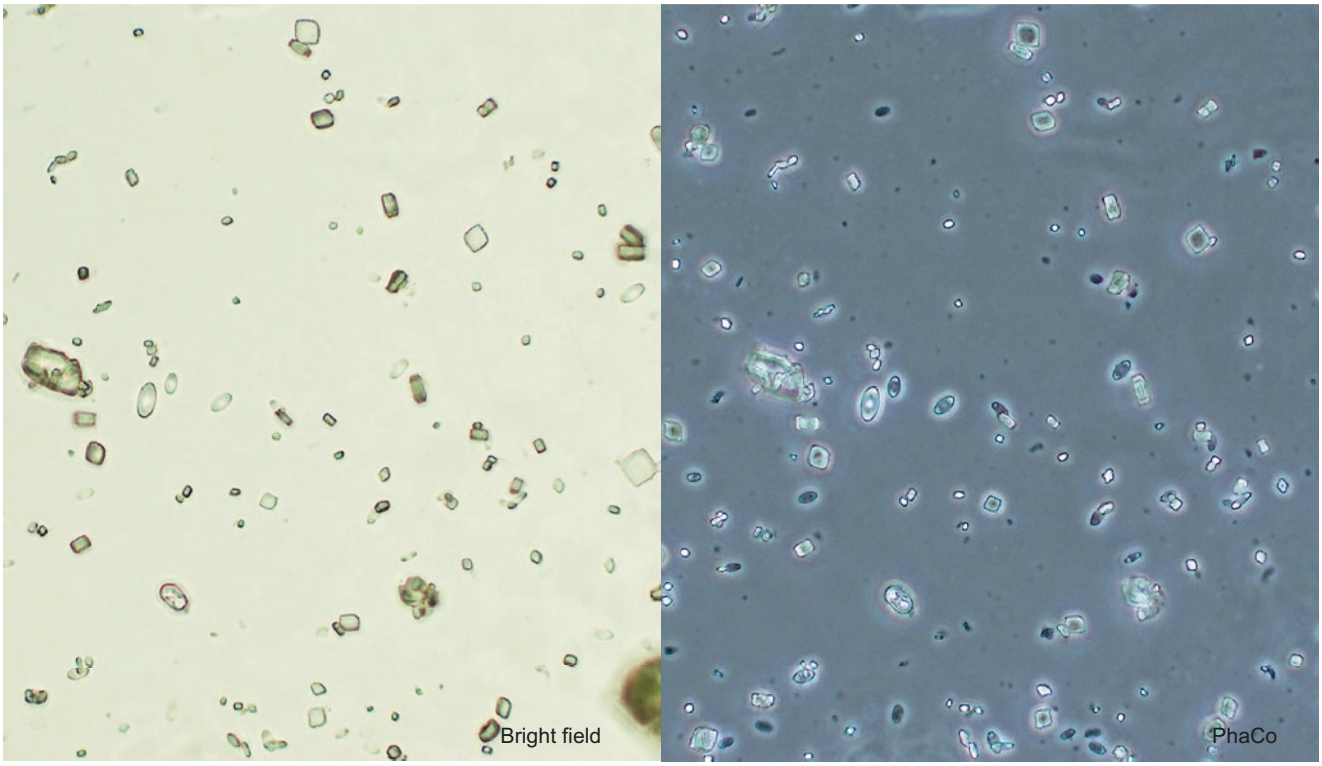
12.4.22 Bacteriuria and Feces



EumEc:	0–1	/HPF	SqEc:	0–1	/HPF
DysEc:		/HPF	Bact:	+++	/HPF
Lc:	0–1	/HPF	Fecal material:	++	/HPF

Fig. 12.63 Bacteriuria and feces

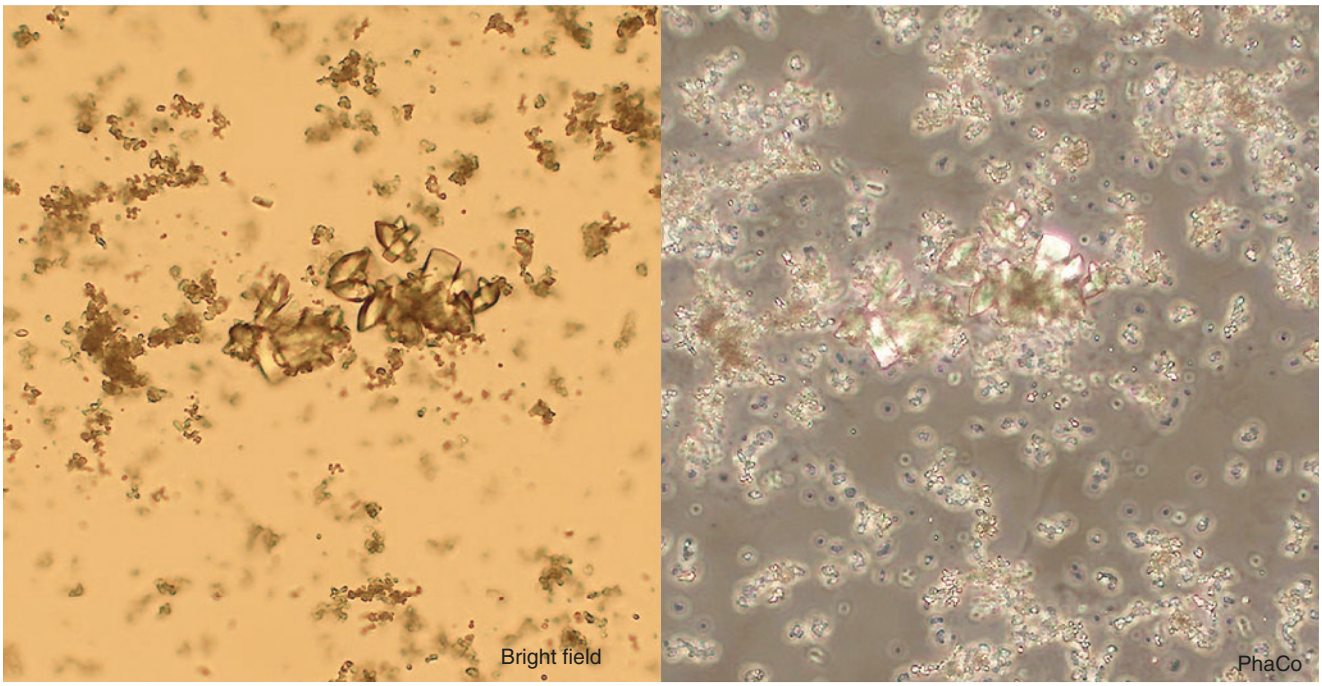
12.4.23 Crystalluria (Uric Acid Crystals and Calcium Oxalates)



EumEc:	0–1	/HPF	SqEc:	0–1	/HPF
DysEc:		/HPF	Bact:	(+)	/HPF
Lc:	0–1	/HPF	Uric acid crystals: (diamond shape)	+	/HPF
			Ca-oxalates: (roundish-oval)	+	/HPF

Fig. 12.64 Crystalluria I: uric acid crystals and calcium oxalates

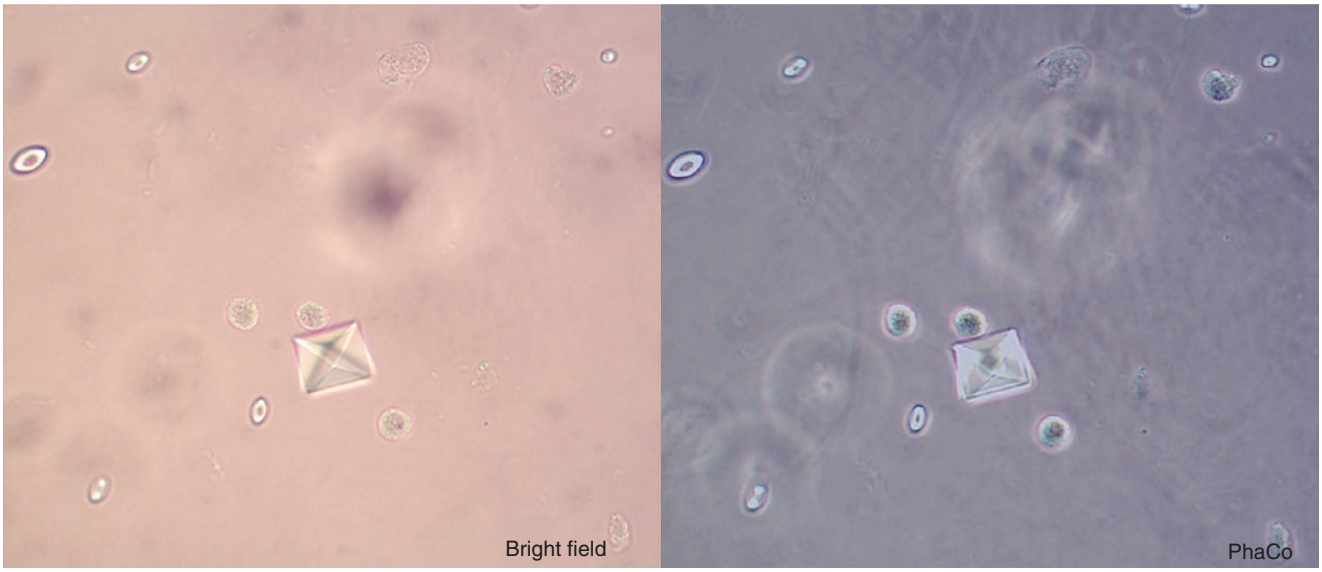
12.4.24 Crystalluria (Uric Acid Crystals and Urates)



EumEc:	0–1	/HPF	SqEc:	0–1	/HPF
DysEc:		/HPF	Bact:	(+)–+	/HPF
Lc:	0–1	/HPF	Uric acid crystals:	+	/HPF
			Urates:	++	/HPF

Fig. 12.65 Crystalluria II: uric acid crystals and urates, urine pH = 5.5

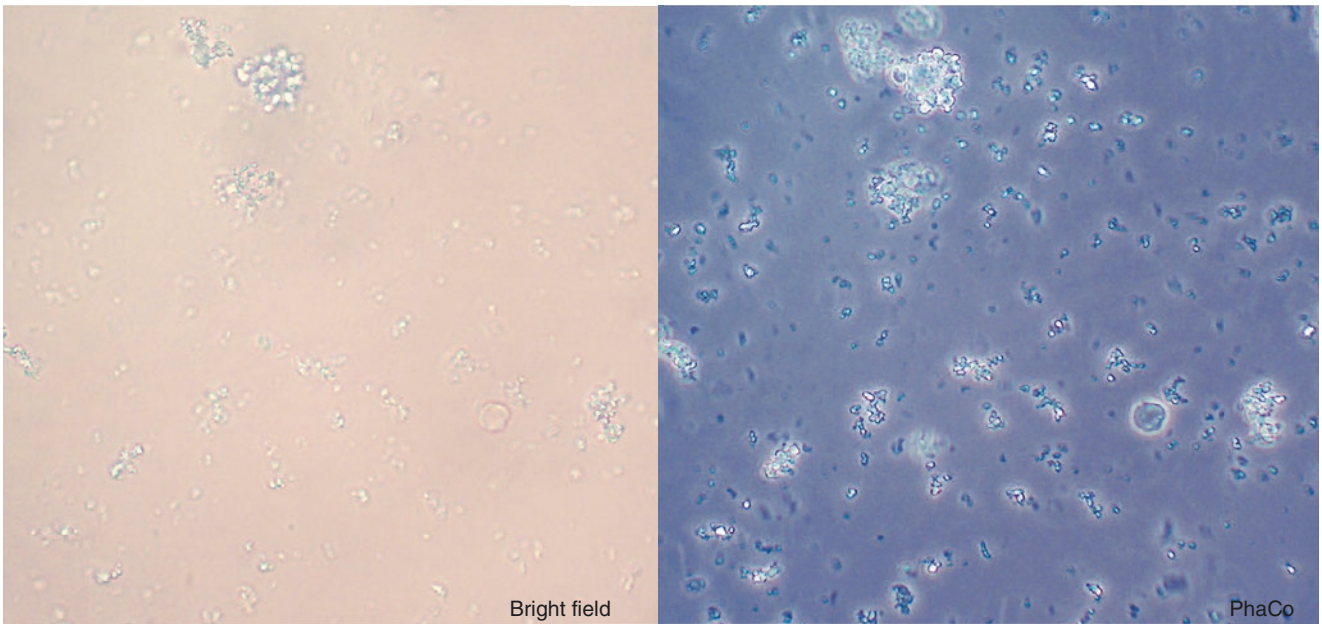
12.4.25 Crystalluria (Square/Envelope-Shaped and Round/Oval Calcium Oxalates)



EumEc:	0–1	/HPF	SqEc:	0–1	/HPF
DysEc:	–	/HPF	Bact:	(+)	/HPF
Lc:	1–4	/HPF	Ca-oxalates:	+	/HPF

Fig. 12.66 Crystalluria III: square/envelope-shaped and round/oval calcium oxalates

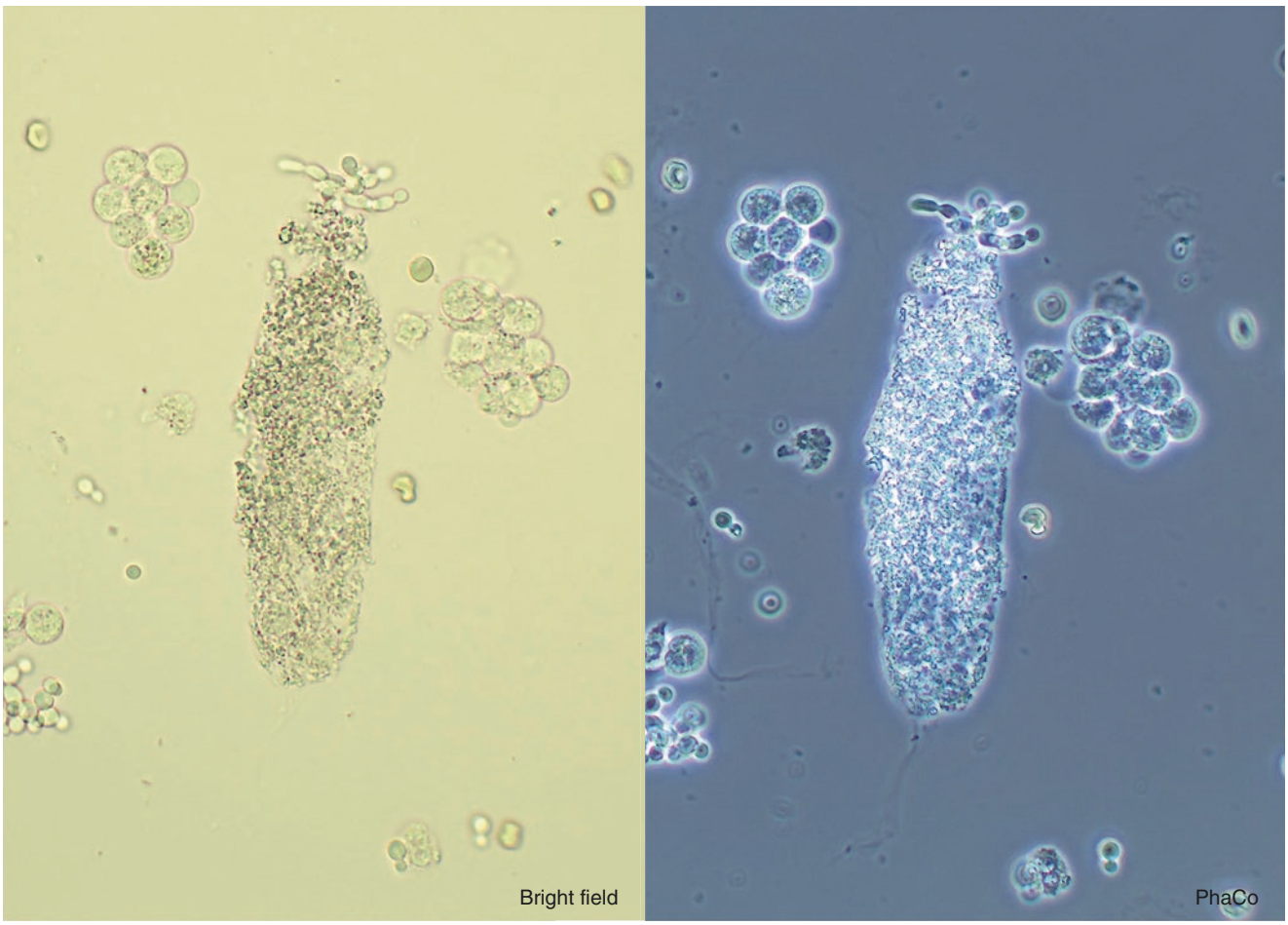
12.4.26 Crystalluria (Amorphous Phosphates)



EumEc:	0–1	/HPF	SqEc:	0–1	/HPF
DysEc:	-	/HPF	Bact:	(+)–+	/HPF
Lc:	0–1	/HPF	Amorphous phosphates:	+	/HPF

Fig. 12.67 Crystalluria IV: amorphous phosphates, urine pH = 8

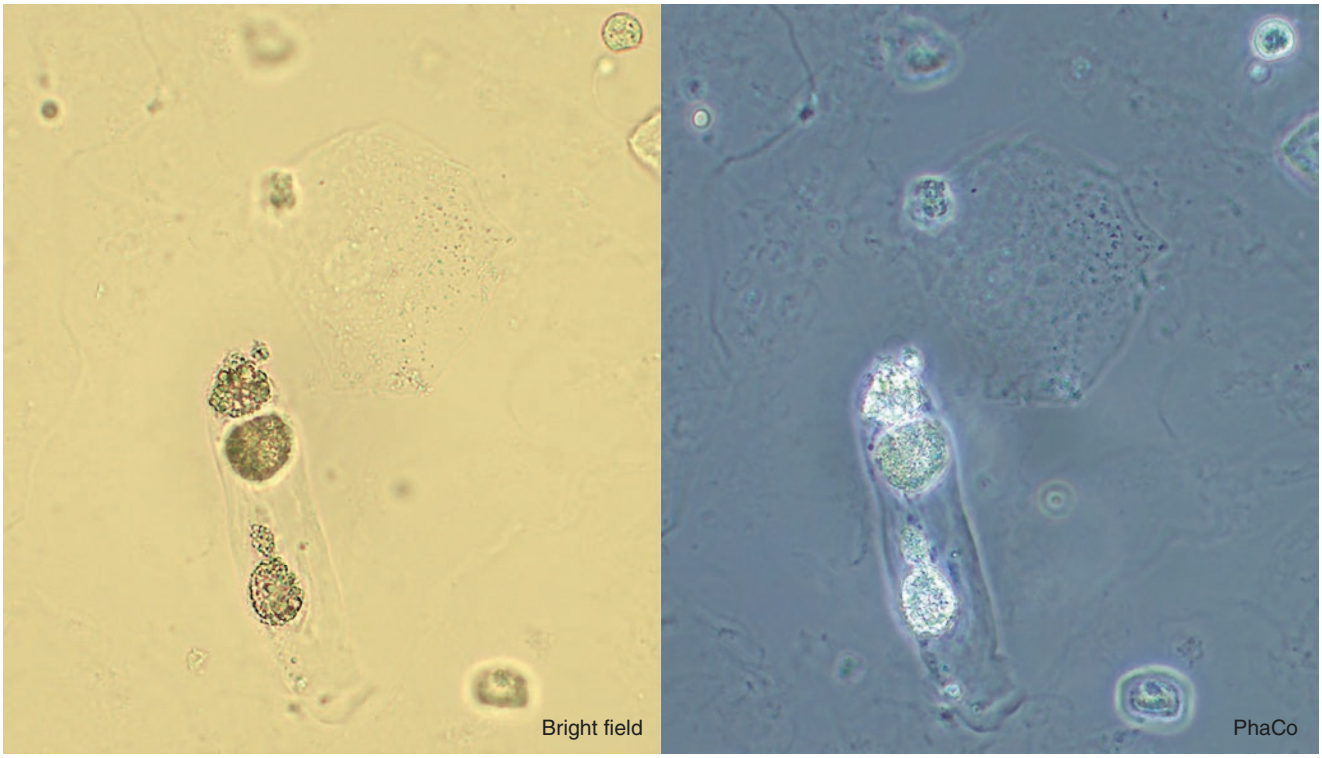
12.4.27 Granular Casts



EumEc:	5–15	/HPF	SqEc:	0–1	/HPF
DysEc:		/HPF	Bact:	(+)	/HPF
Lc:	5–15	/HPF	Yeast cells:	(+)	/HPF
			GranCa:	3	/aHPF

Fig. 12.68 Granular casts

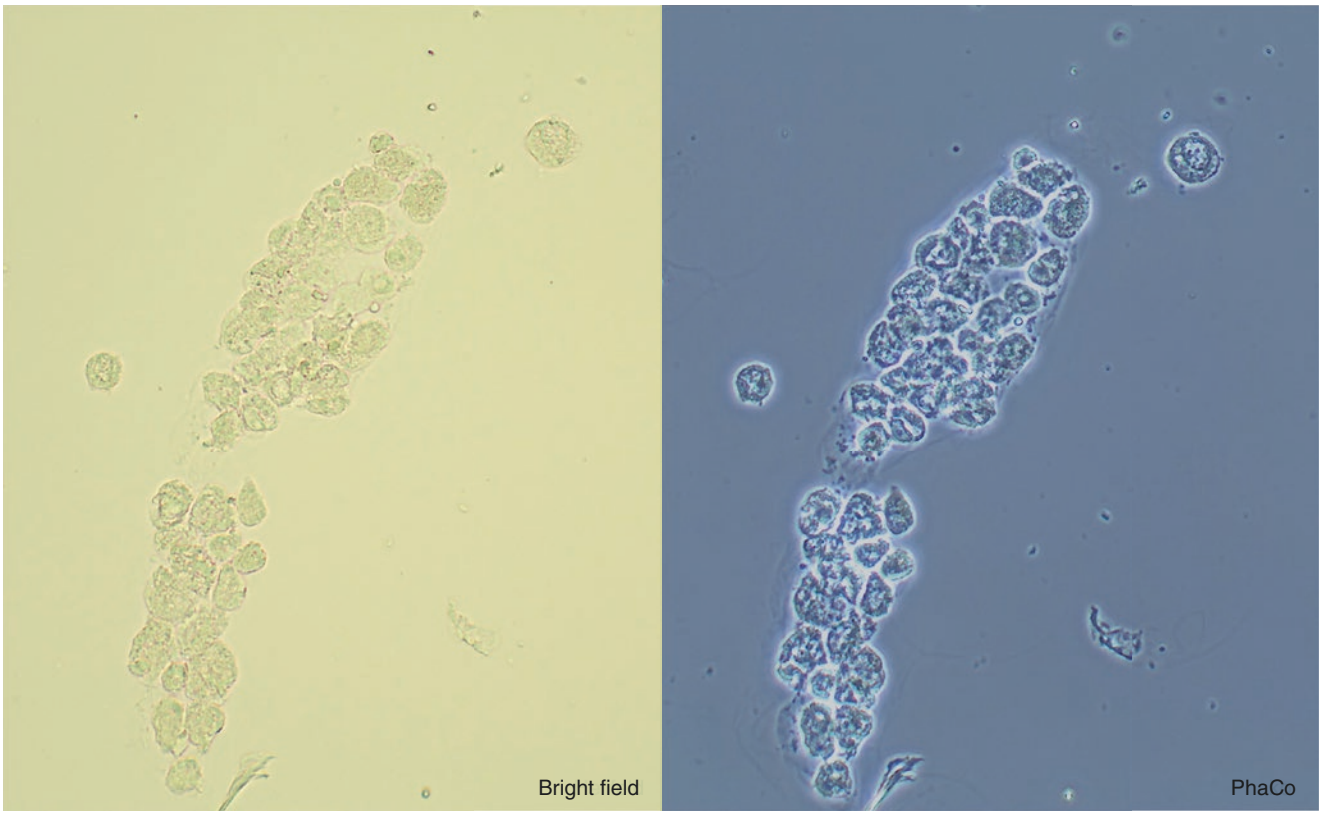
12.4.28 Lipiduria with Oval Fat Body Casts



EumEc:	0–1	/HPF	SqEc:	0–1	/HPF
DysEc:		/HPF	Bact:	(+)	/HPF
Lc:	0–1	/HPF	OFB casts:	7	/aHPF

Fig. 12.69 Oval fat body casts

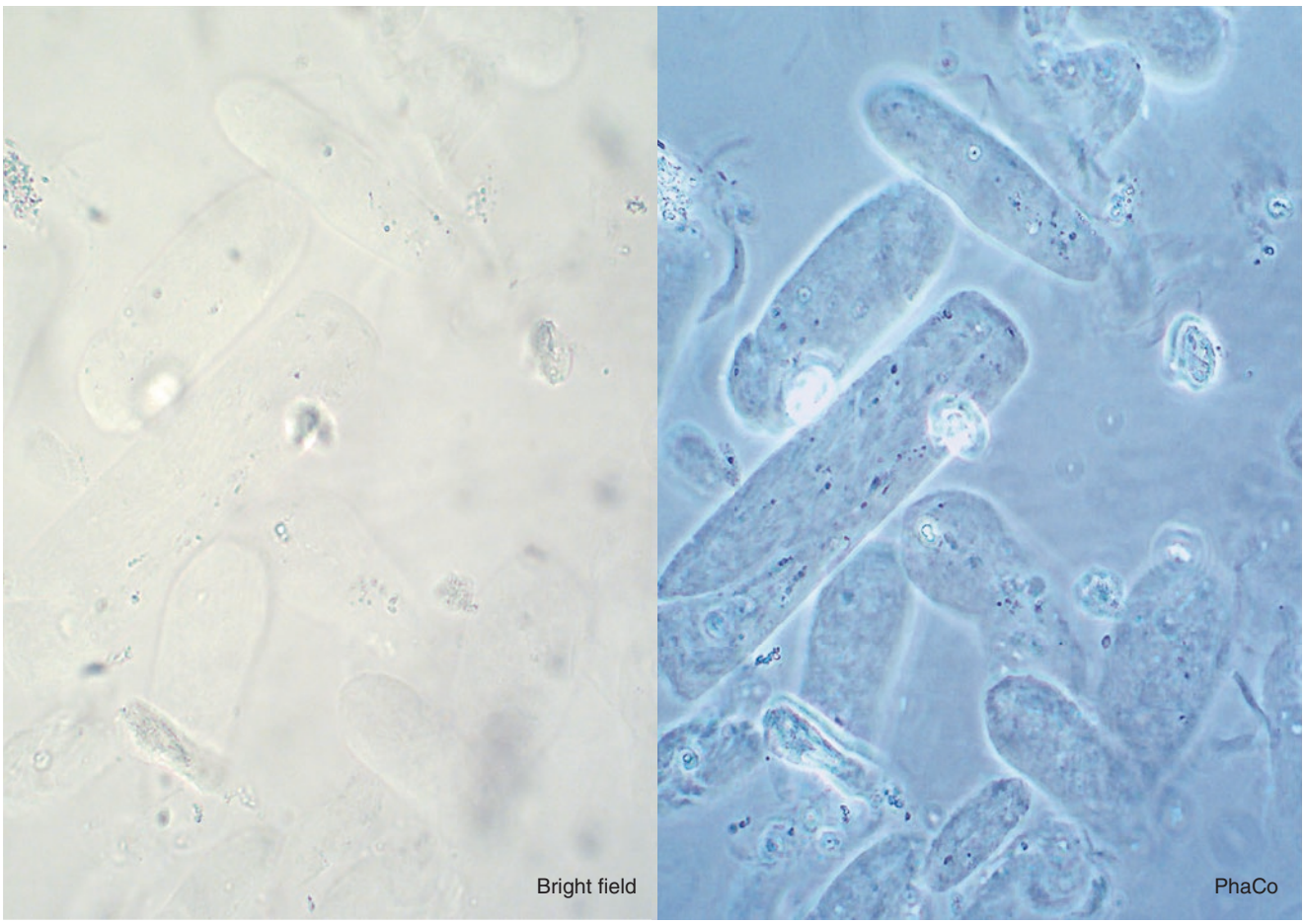
12.4.29 Epithelial Casts



EumEc:	0–1	/HPF	SqEc:	0–1	/HPF
DysEc:		/HPF	Bact:	(+)	/HPF
Lc:	0–1	/HPF	Epithelial casts:	10	/aHPF
Renal epithelial cells:	0–1	/HPF			

Fig. 12.70 Epithelial casts

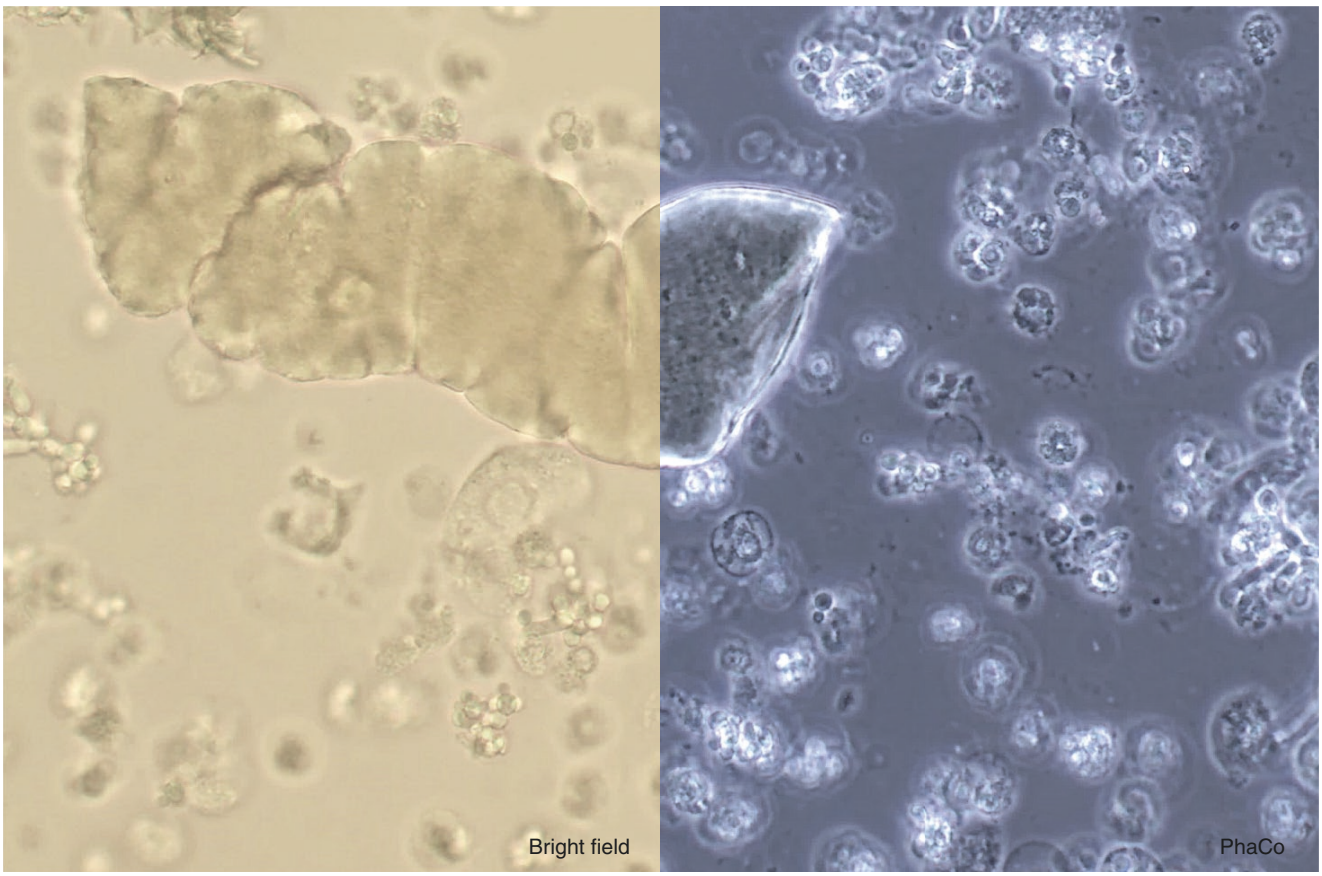
12.4.30 Cylindruria (Hyaline Casts)



EumEc:	0–1	/HPF	SqEc:	0–1	/HPF
DysEc:		/HPF	Bact:	(+)	/HPF
Lc:	0–1	/HPF	HyalCa:	110	/aHPF

Fig. 12.71 Cylinduria (hyaline casts)

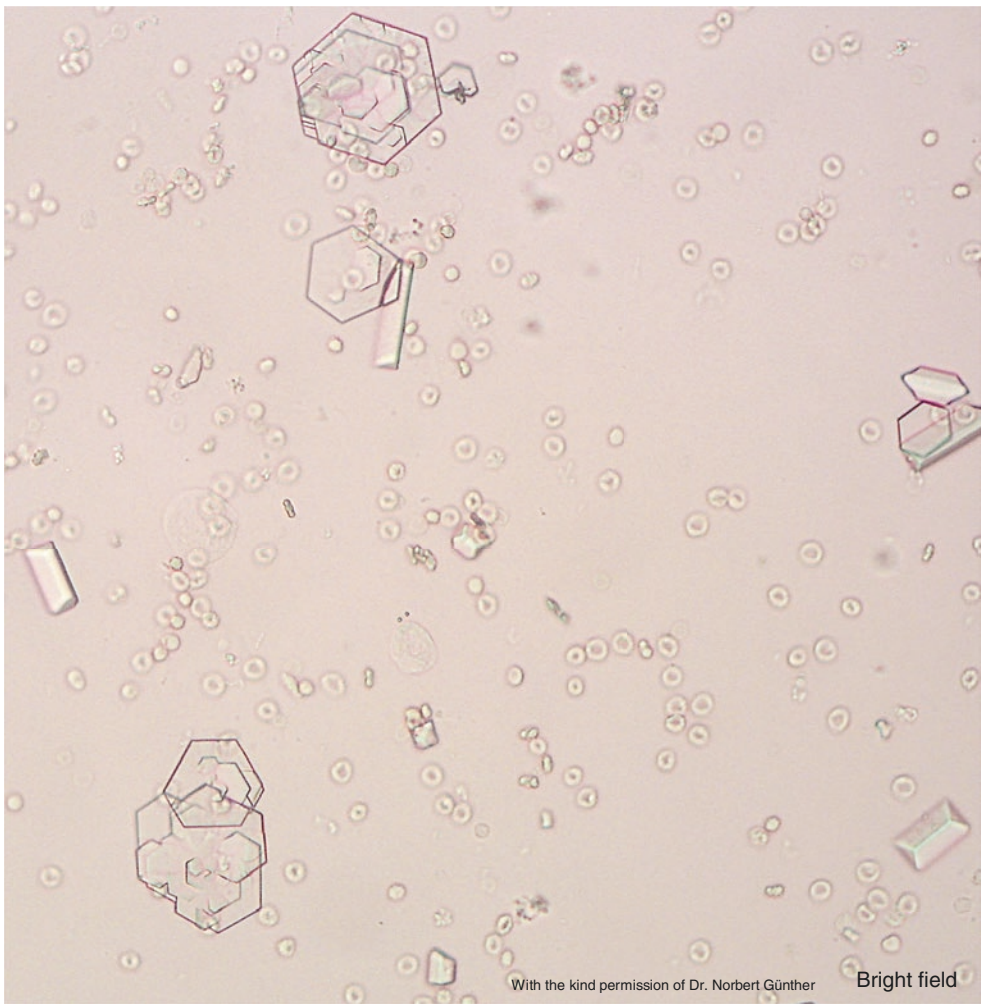
12.4.31 Waxy Cast, Leukocyturia, and Yeast Cells



EumEc: 0–1	/HPF	SqEc: 0–1	/HPF
DysEc:	/HPF	Bact: (+)–+	/HPF
Lc: 15–50	/HPF	Yeast cells: +	/HPF
Transitional epithelial cells: 0–1	/HPF	Waxy cast: 1	/aHPF

Fig. 12.72 Waxy Cast, leukocyturia, and yeast cells

12.4.32 Cystinuria and Eumorphic Hematuria



EumEc:	>50	/HPF	SqEc:	0–1	/HPF
DysEc:		/HPF	Bact:	+	/HPF
Lc:	1–4	/HPF	Cystine:	+	/HPF
Transitional epithelial cells: 1–4		/HPF	Triple phosphates:	+	/HPF

Fig. 12.73 Cystinuria and eumorphic hematuria

Part IV

Urine Sediment Quiz

13

13.1 Quiz on Urinary Sediment Constituents

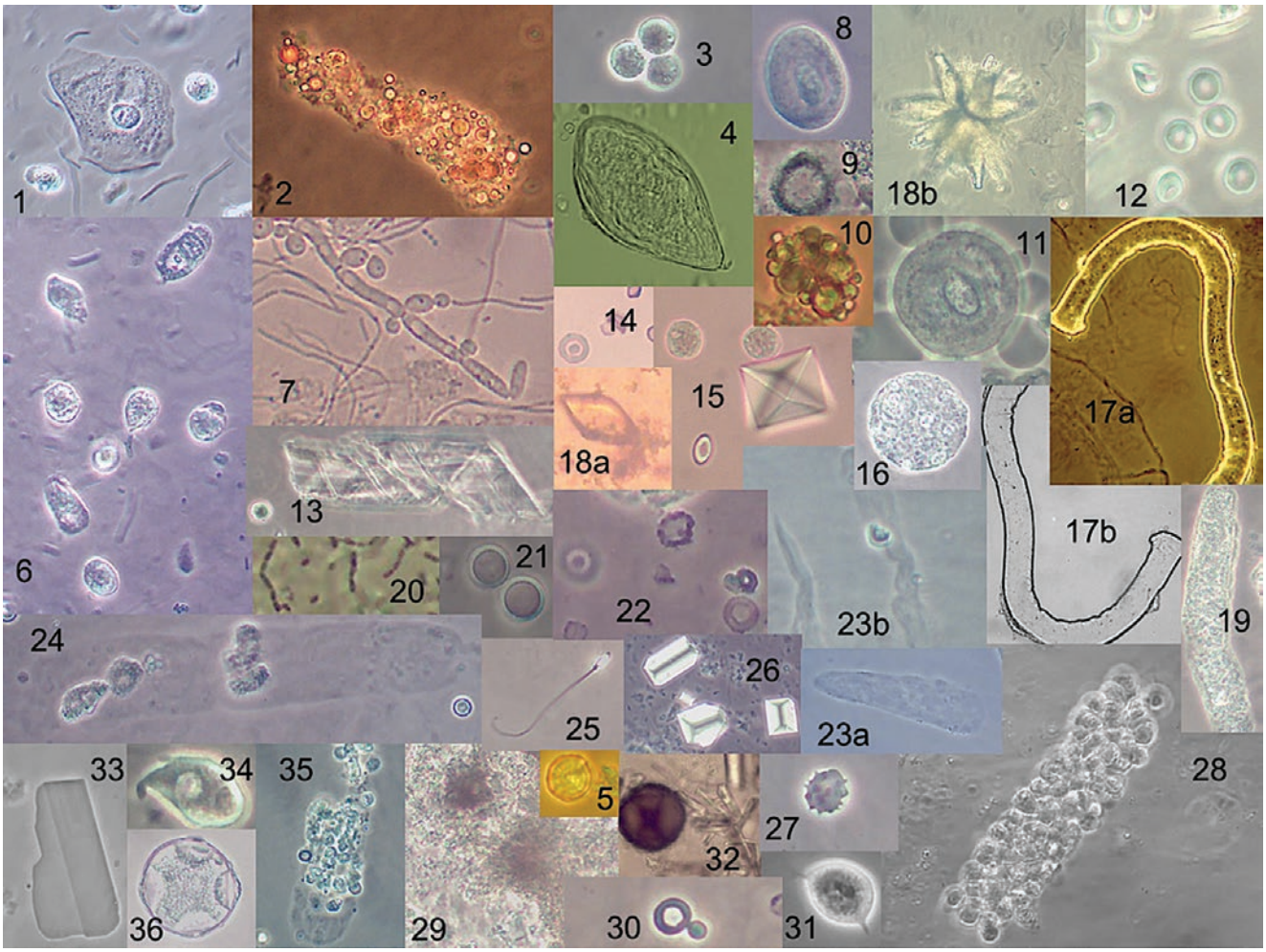


Fig. 13.1 Quiz on urinary sediment constituents

J. Neuendorf, *Urine Sediment*, https://doi.org/10.1007/978-3-030-15911-5_13

13.2 Quiz on Urinary Sediment Constituents: Answers

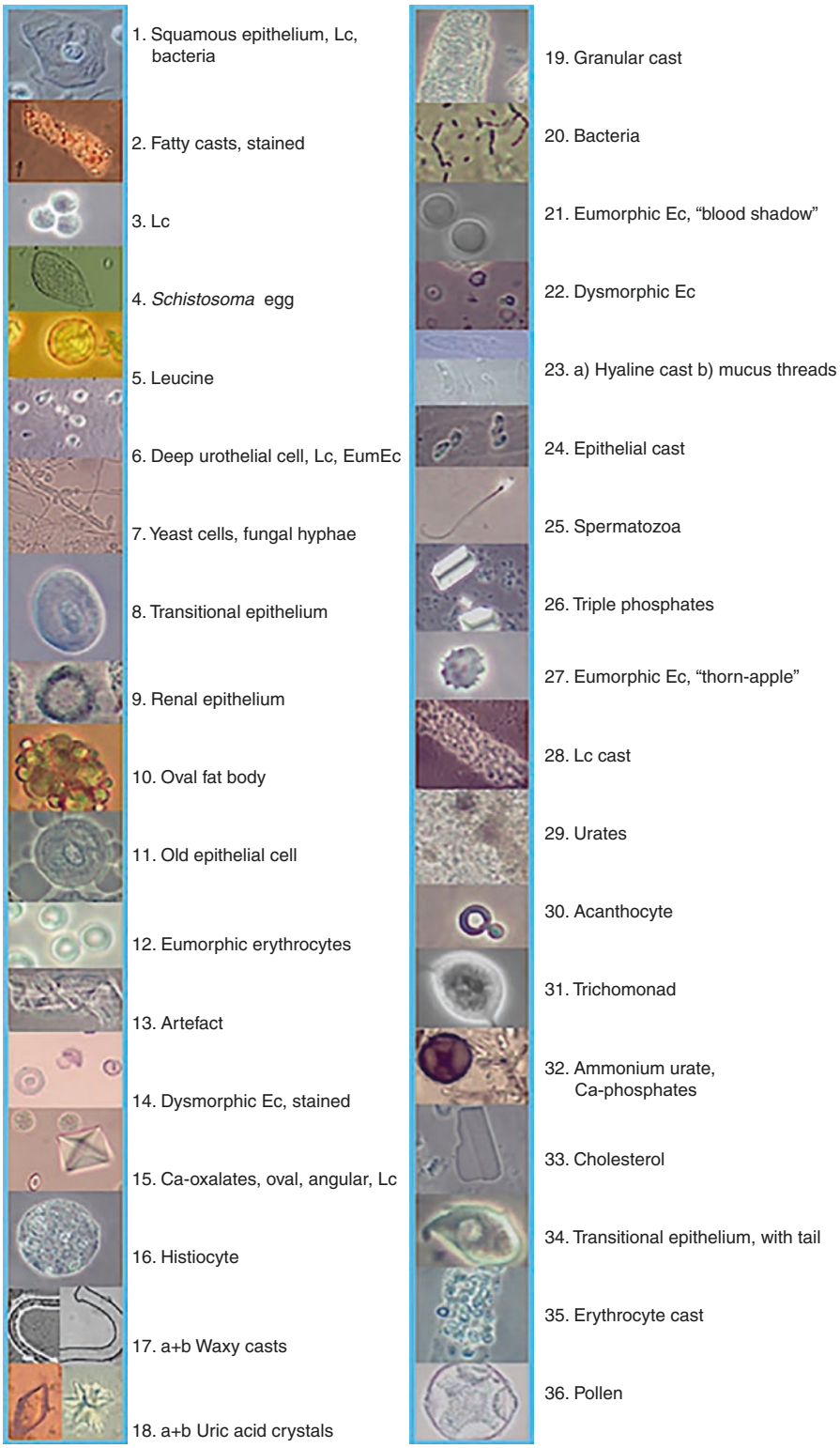


Fig. 13.2 Quiz on urinary sediment constituents: answers

13.3 Exercise Sheet to Fill Out

Fig. 13.3 Exercise sheet to fill out

13.4 What Is What? Bacteriuria and/or Crystalluria?

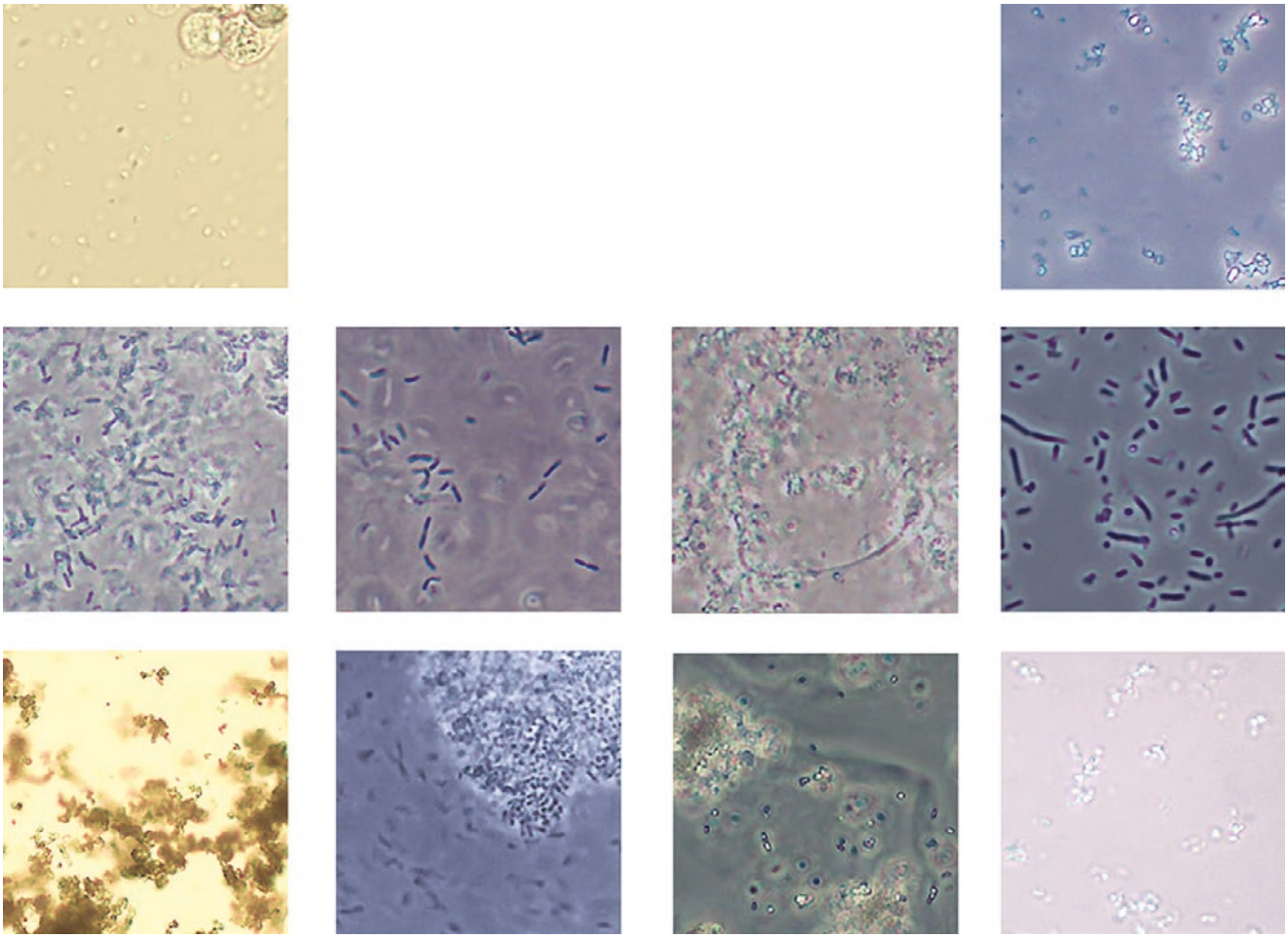


Fig. 13.4 What is what? Bacteriuria and/or crystalluria?

13.4.1 Answers

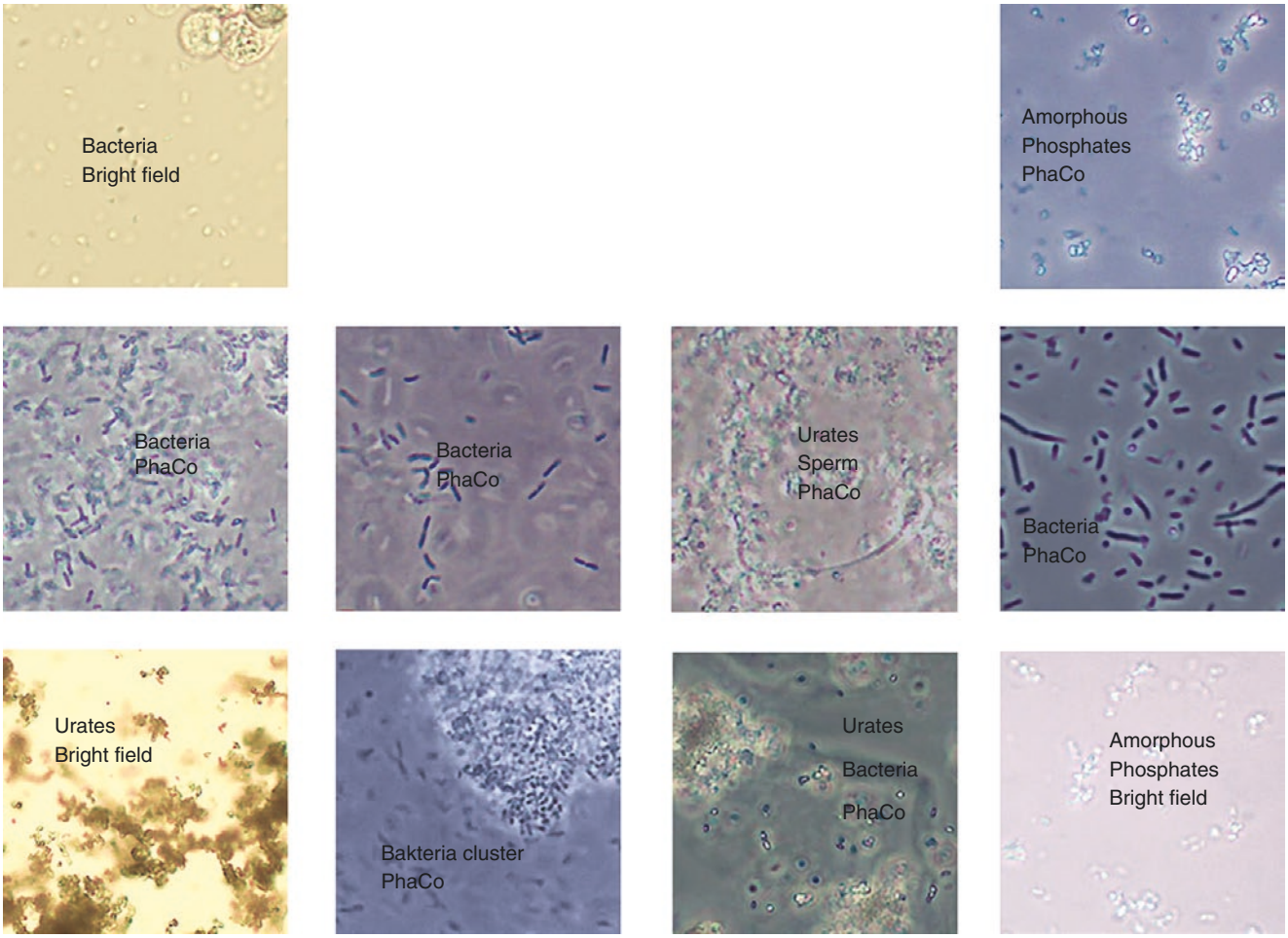


Fig. 13.5 What is what? Bacteriuria and/or crystalluria?

13.5 What Is What? Hematuria?

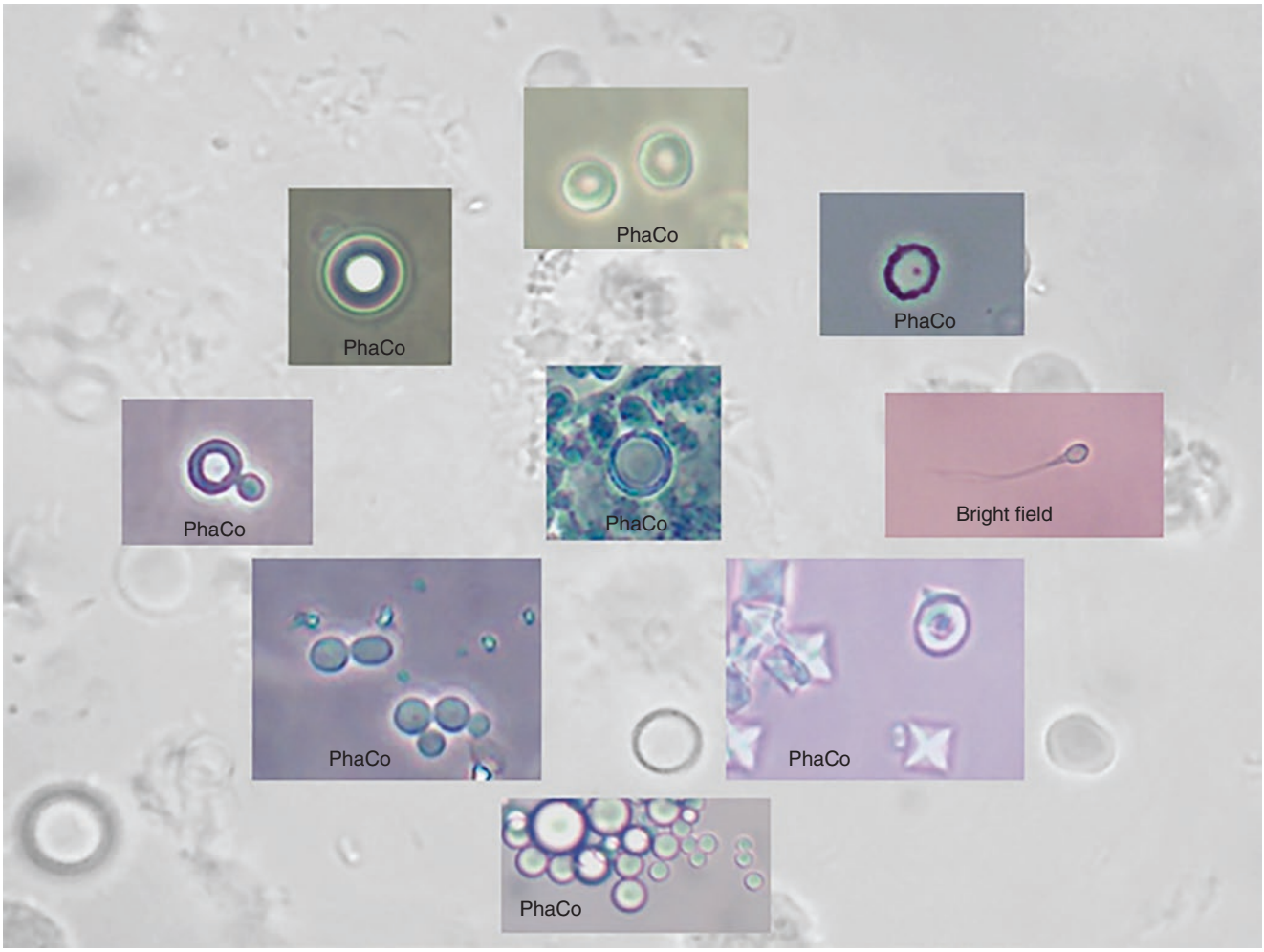


Fig. 13.6 What is what? Hematuria?

13.5.1 Answers

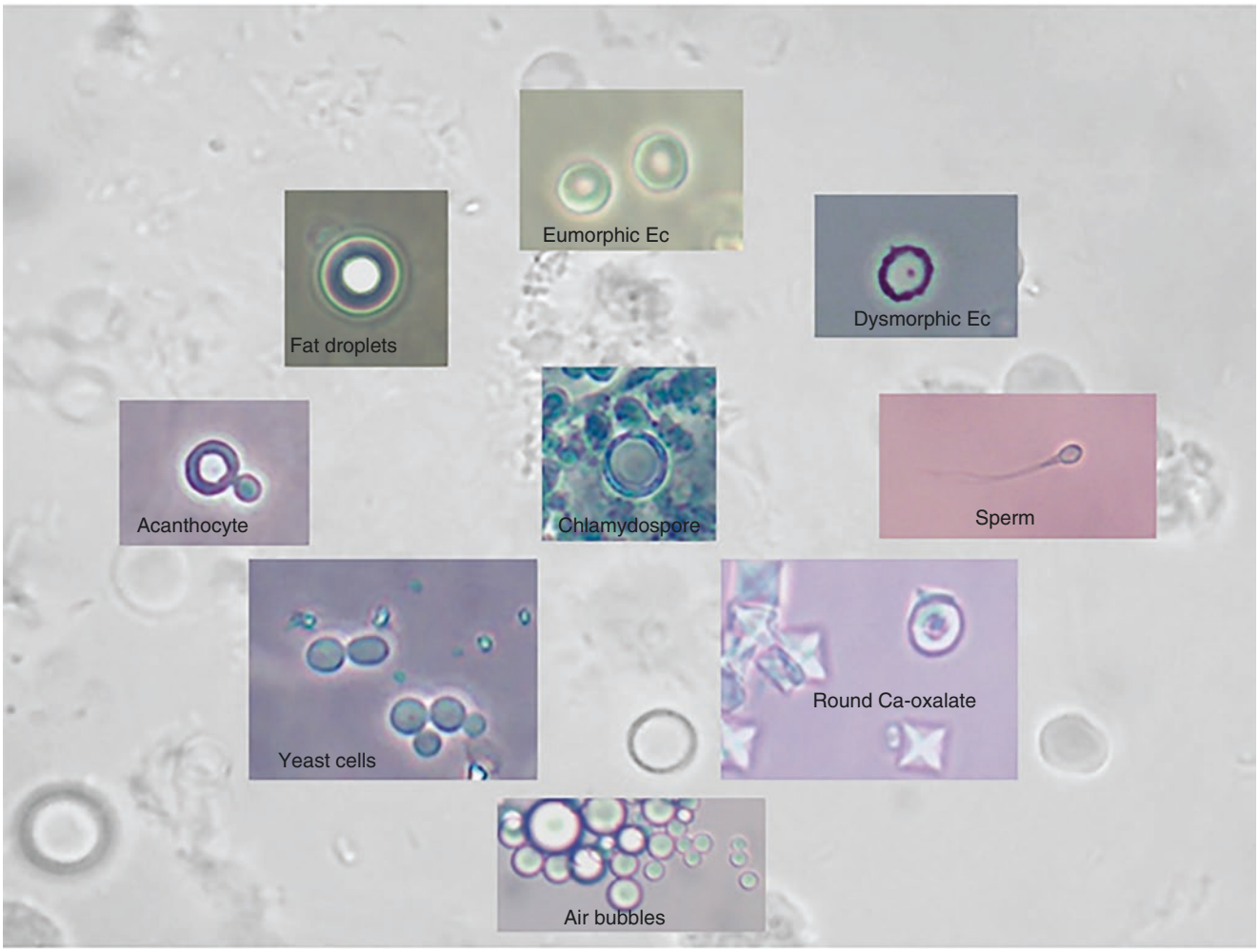


Fig. 13.7 Answers: what is what? Hematuria?

13.6 What Is What?

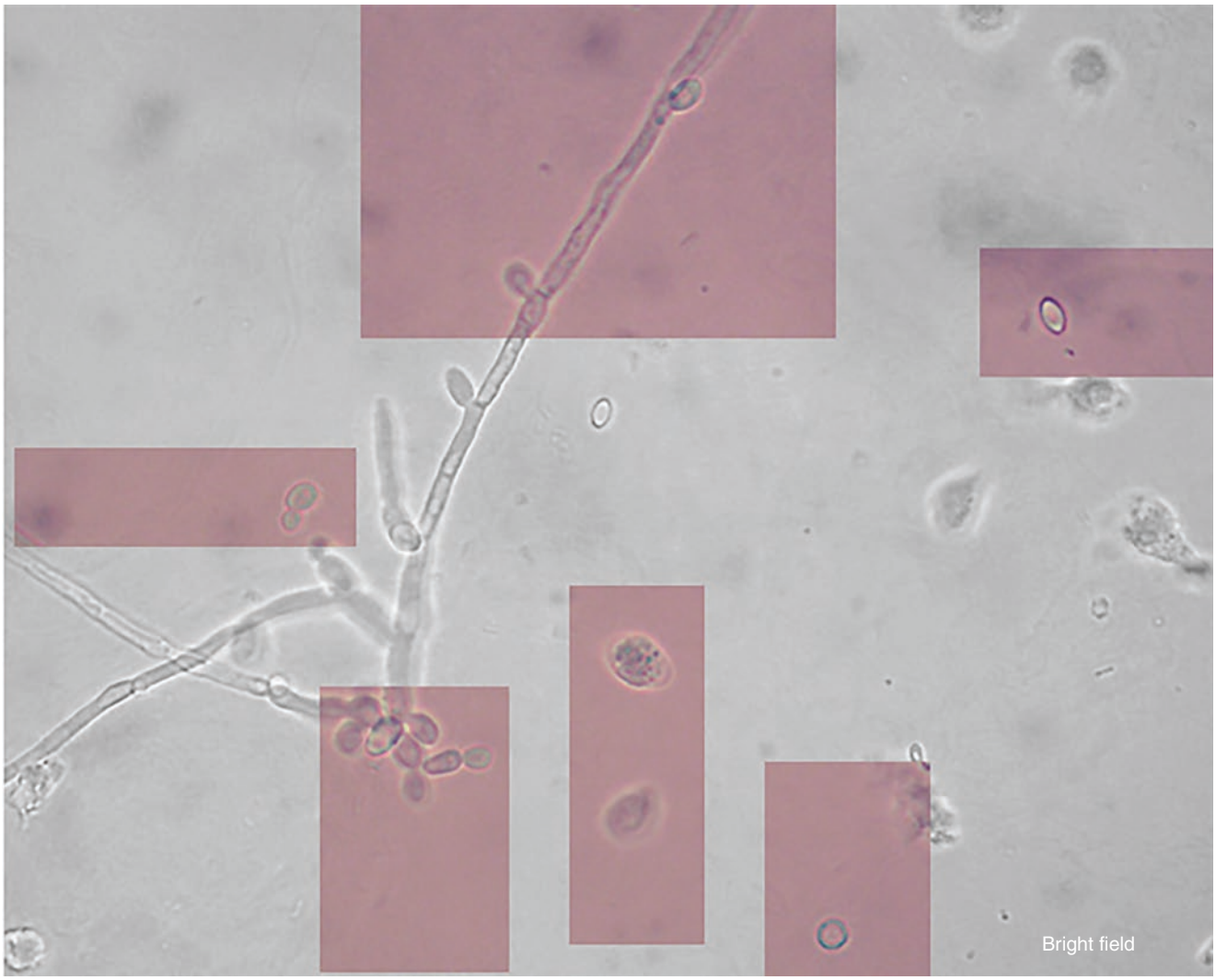


Fig. 13.8 What is what?

13.6.1 Answers

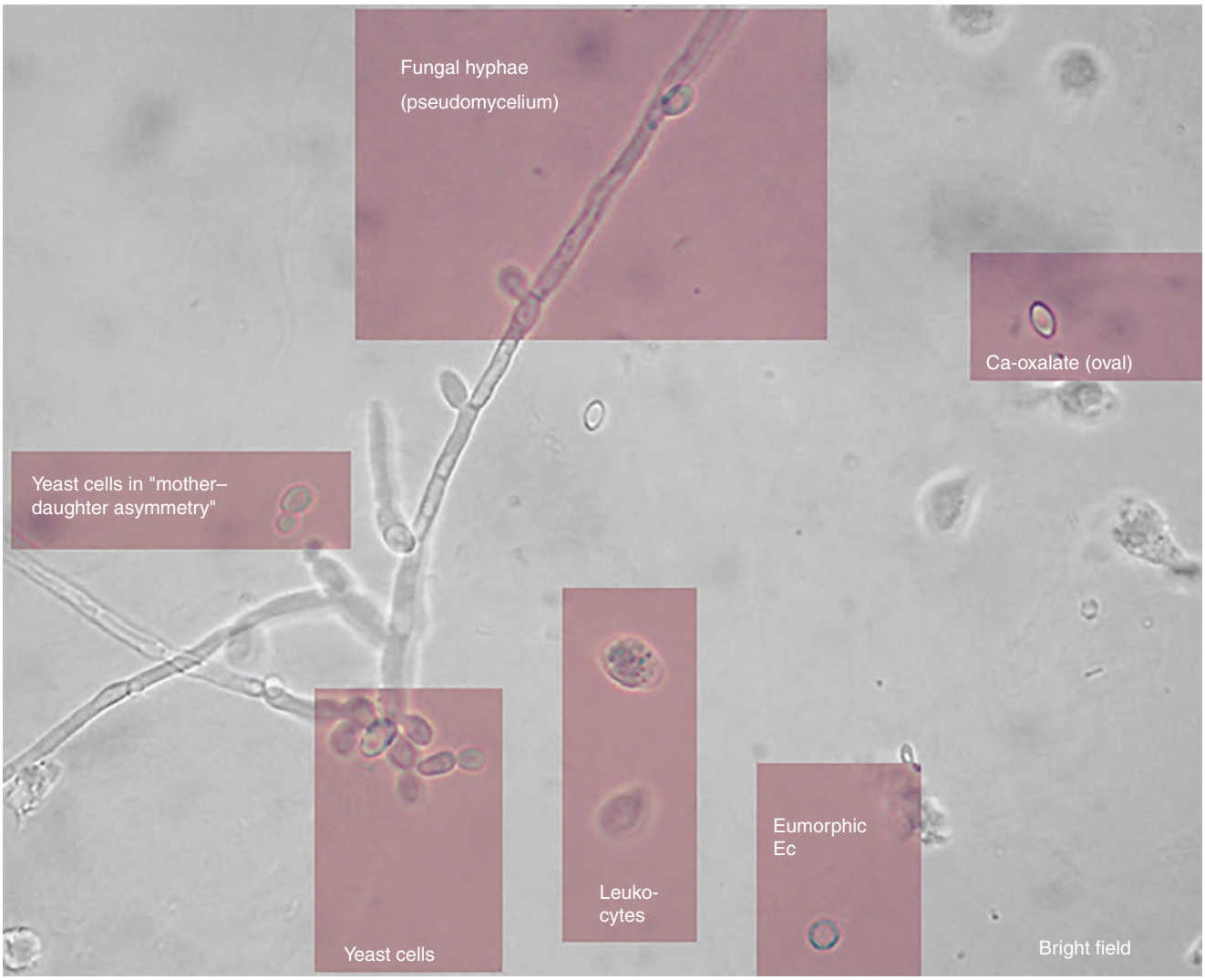


Fig. 13.9 Answers: yeast cells, fungal hyphae, Ca-oxalates, eumorphic erythrocyte, leukocytes

13.7 What Is What?

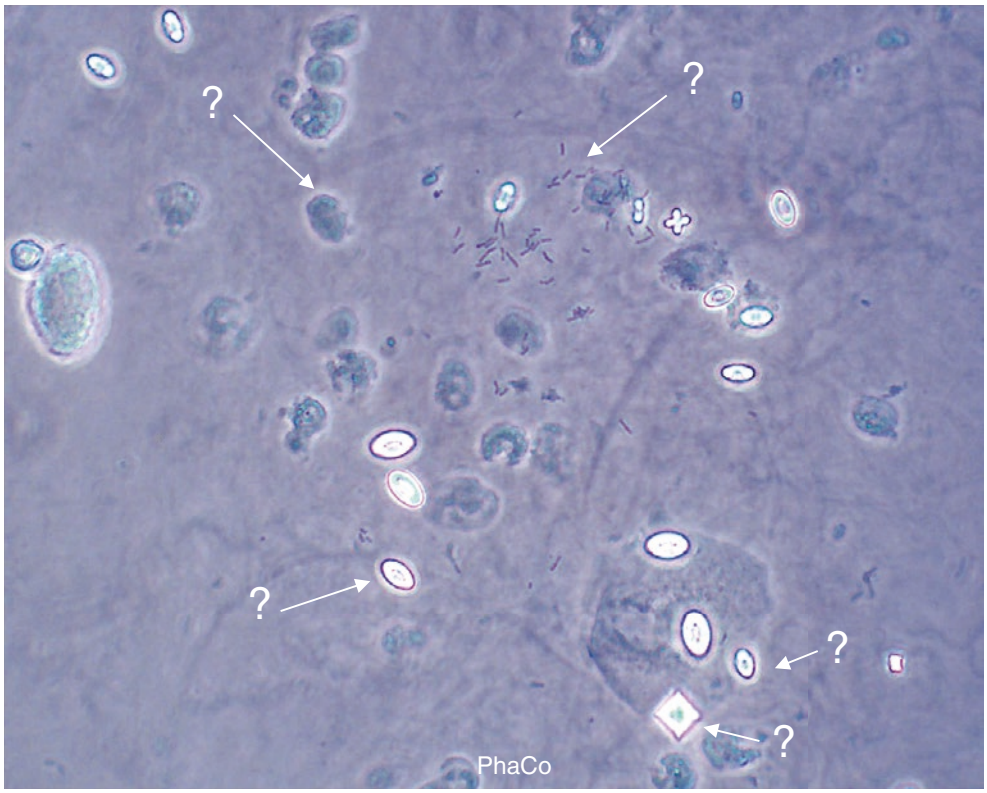


Fig. 13.10 What is what?

13.7.1 Answers

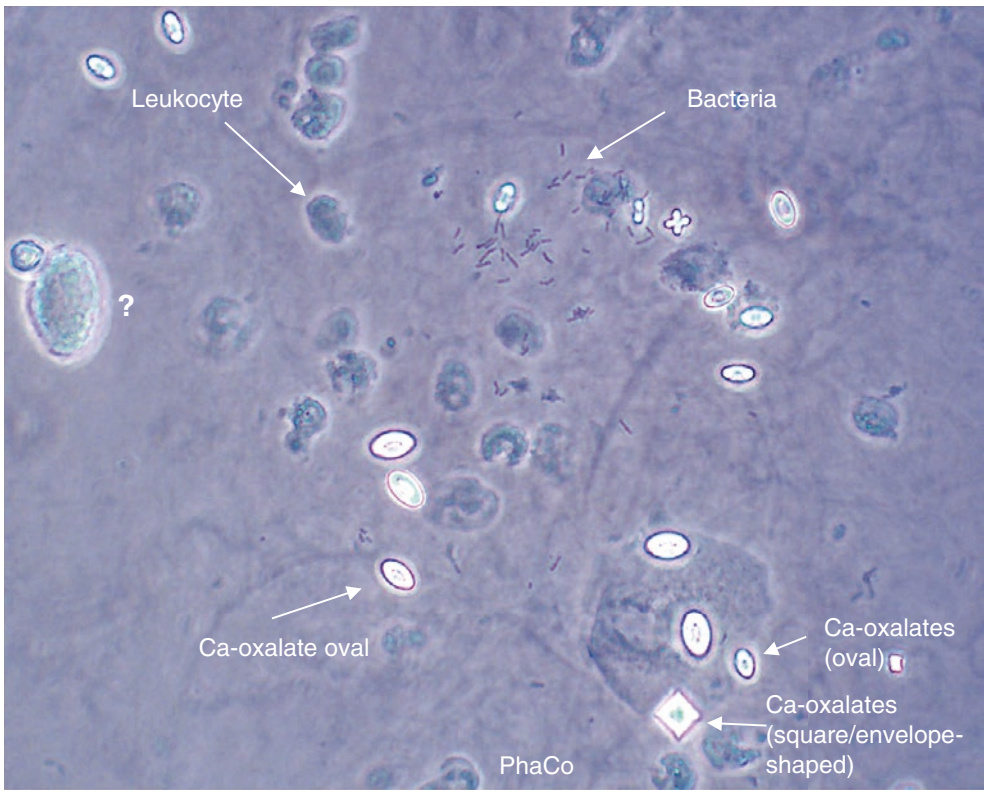


Fig. 13.11 Answers: leukocytes, bacteria, Ca-oxalates

13.8 What Is What?

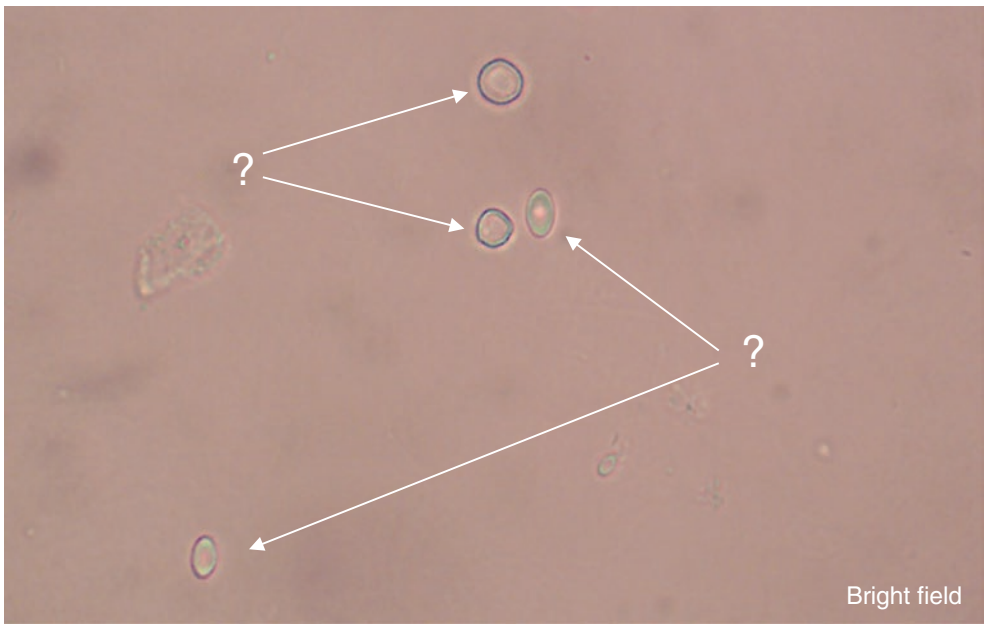


Fig. 13.12 What is what?

13.8.1 Answers

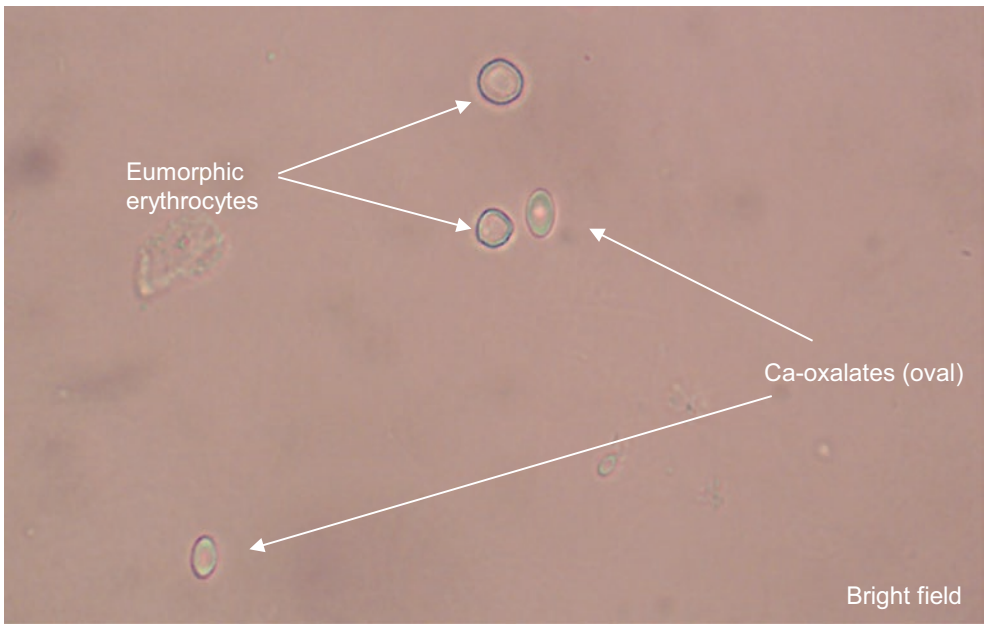
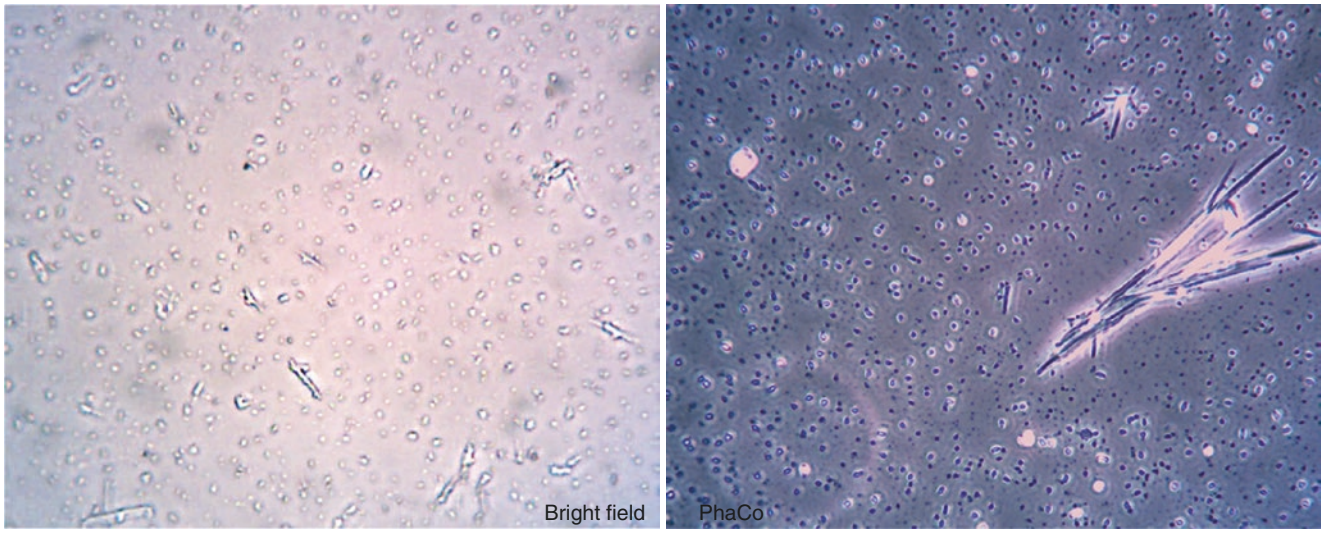


Fig. 13.13 Answer: eumorphic erythrocytes, Ca-oxalates (oval)

13.9 Is the Microscope Plane Correct?



Incorrect plane!

Slides lay on a freshly disinfected work table.

Fig. 13.14 Incorrect plane

13.9.1 Answer

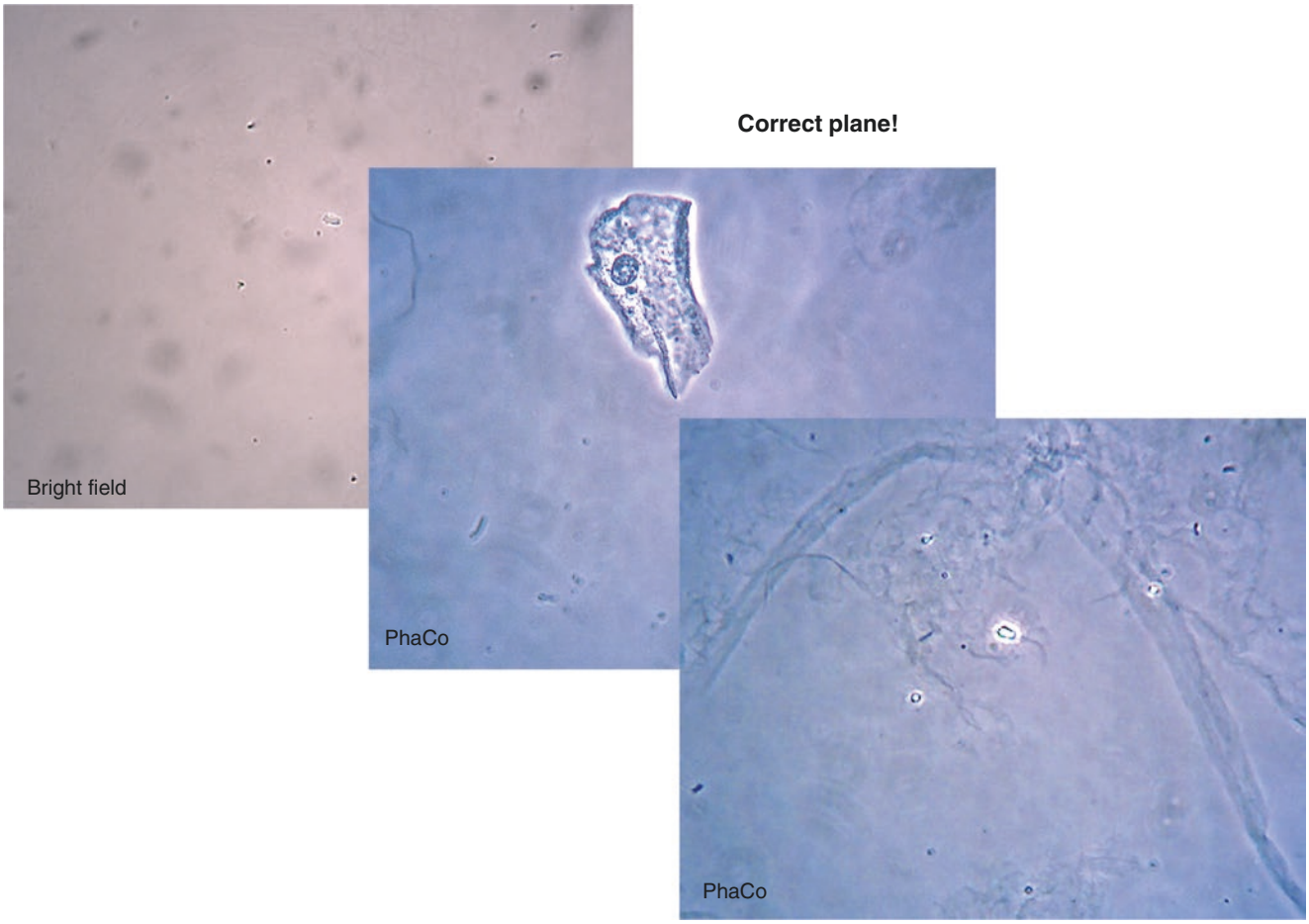


Fig. 13.15 Correct plane

13.10 Schematic Images of Urine Sediment: Quiz

13.10.1 Cellular Constituents, etc.

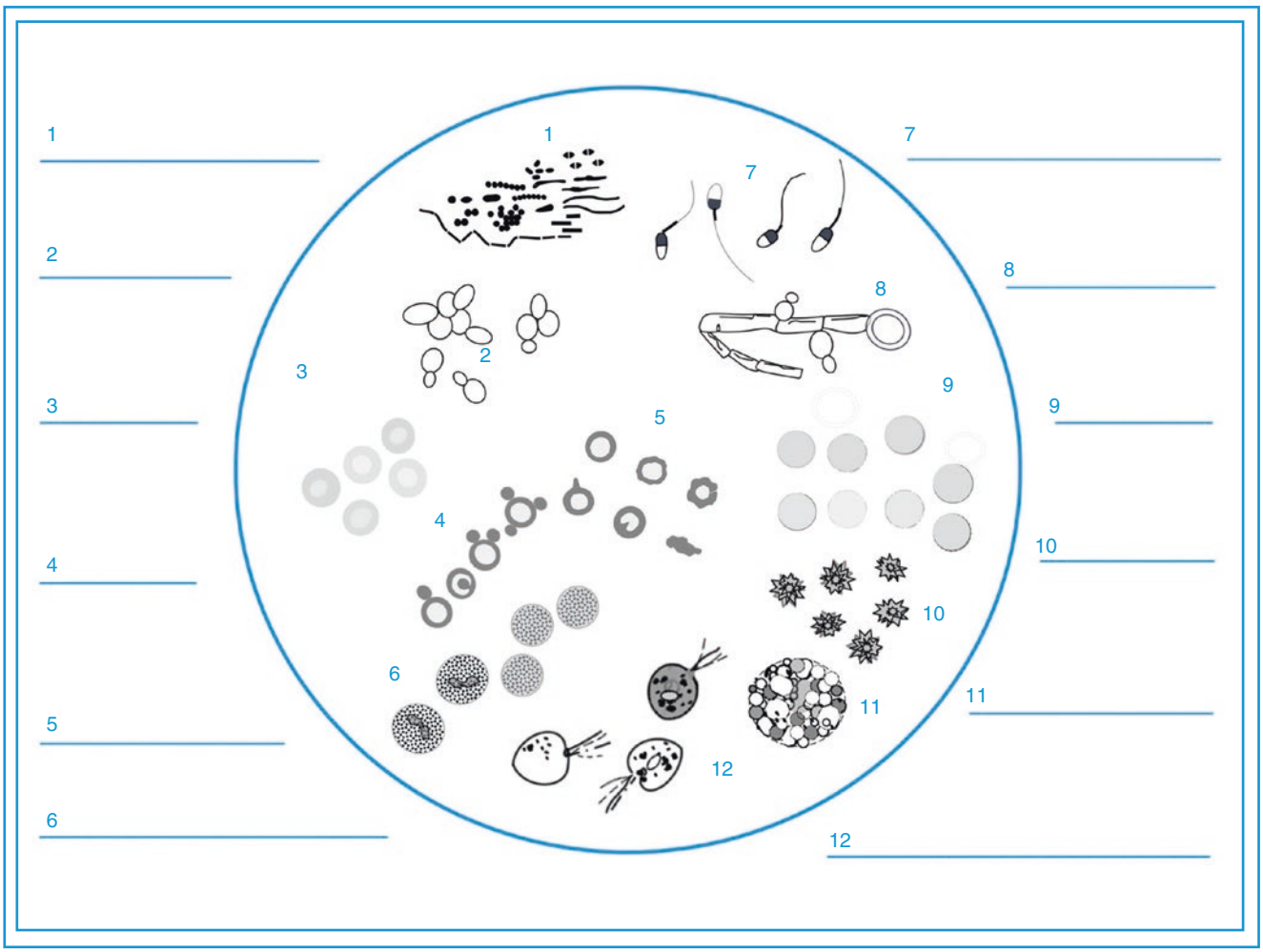


Fig. 13.16 Cellular constituents, etc.

13.10.2 Epithelial Cells

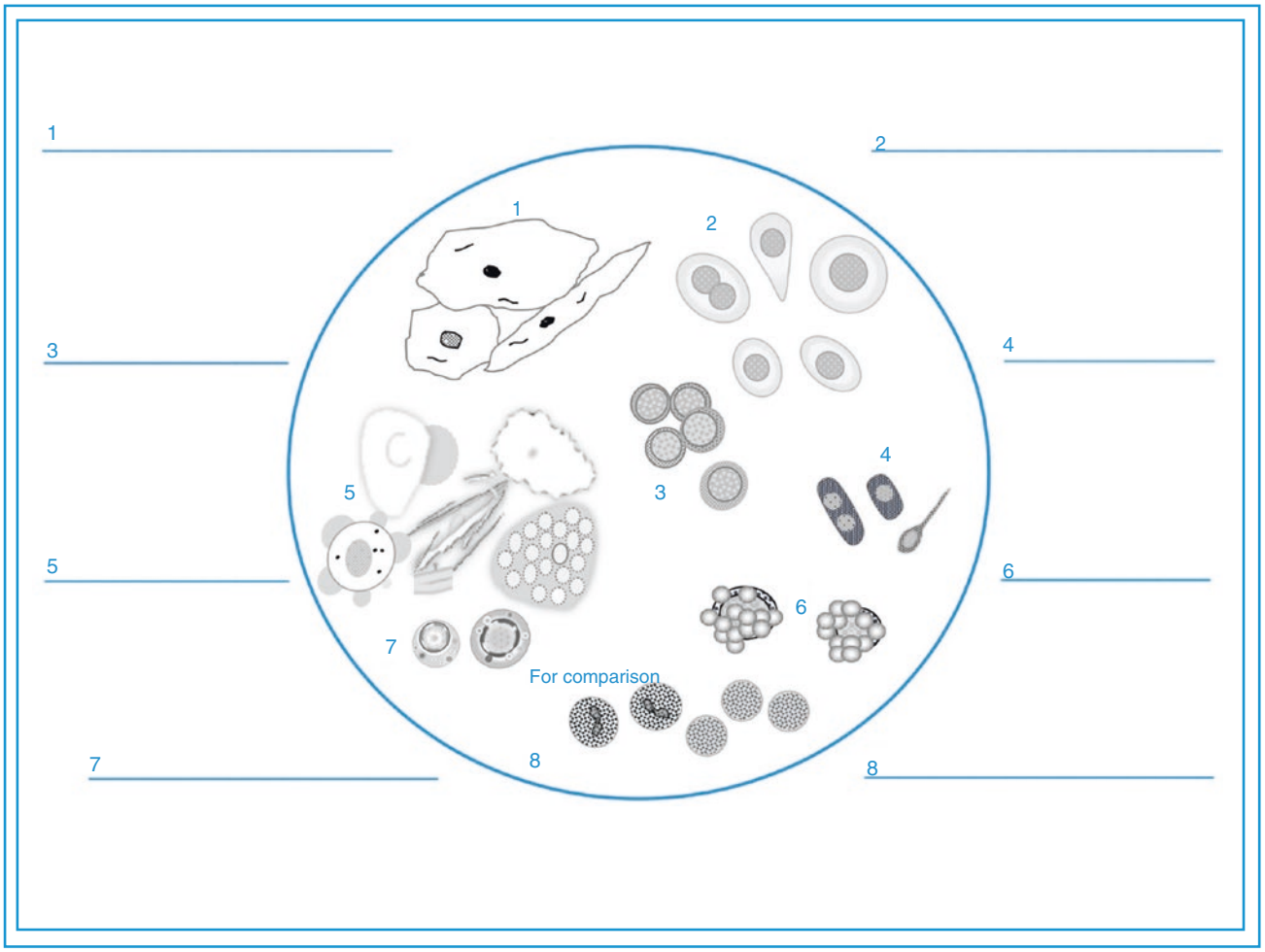


Fig. 13.17 Epithelial cells

13.10.3 Casts

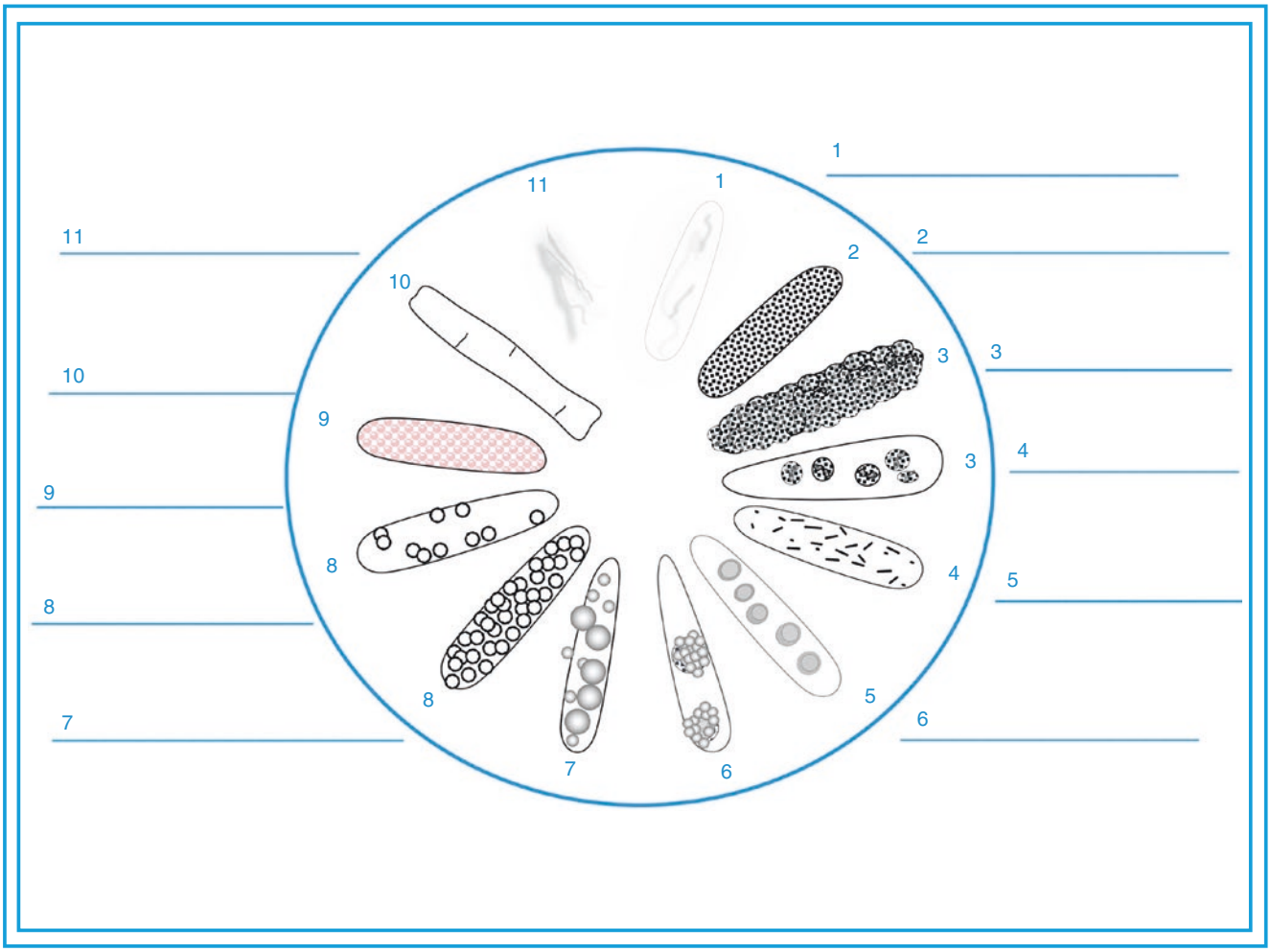


Fig. 13.18 Casts

13.10.4 Crystals

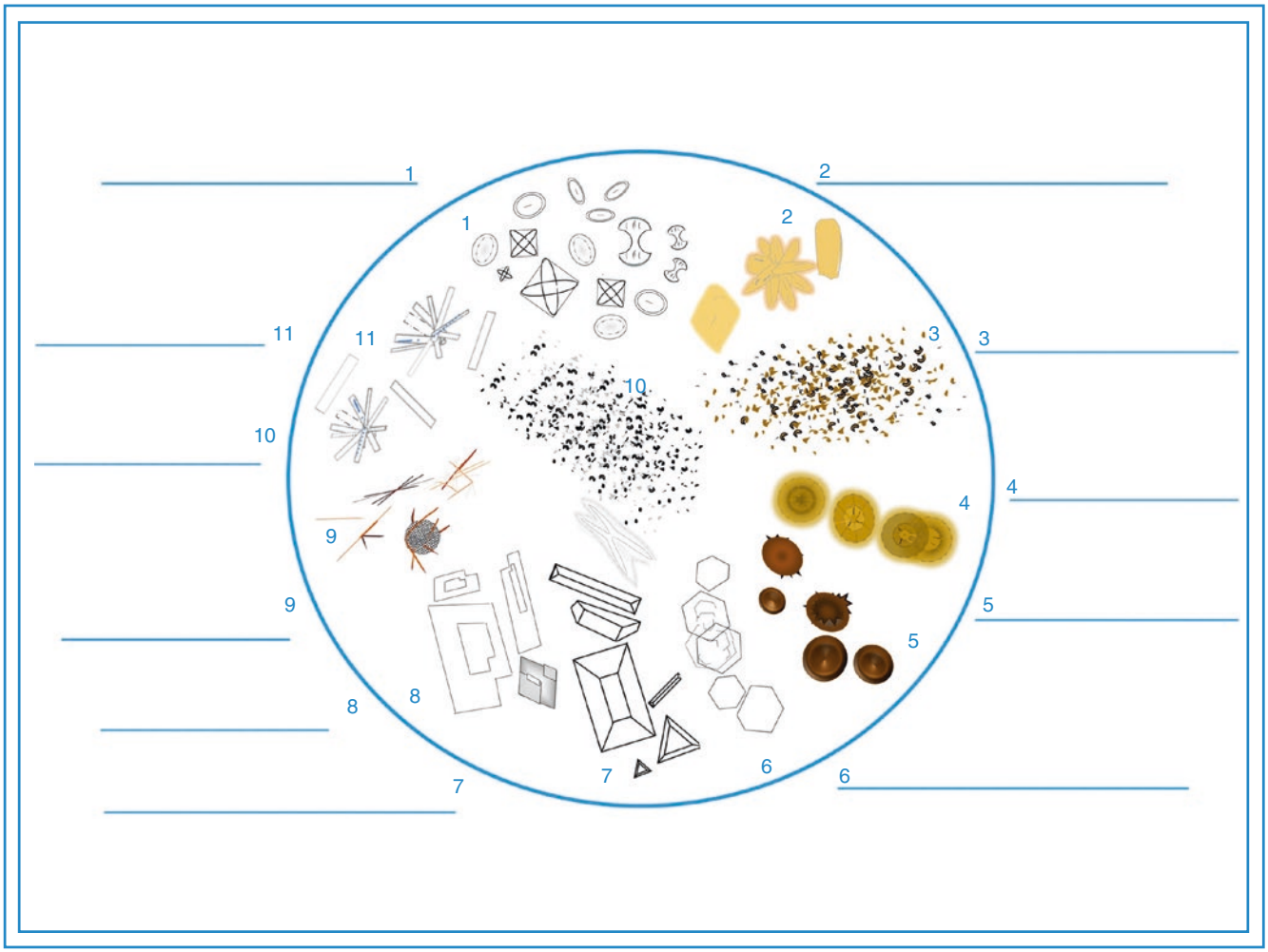


Fig. 13.19 Crystals

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